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Superintendencia de Industria y Comercio
E. S. D.

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO



No. 07-053905- -00005-0000

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Act. 444 RESPUESTAREQUE Folios: 49

Trámite : 011
Evento : 378
Actuación : 444

Referencia : Expediente No. 07053905
Asunto : Solicitud de patente para **CLORHIDRATO DIHIDRATO DE (S)-(-)-1-(4-FLUOROISOQUINOLIN -5-IL)SULFONIL-2-METIL-1,4-HOMOPIPERAZINA**
Solicitante : **KOWA CO., LTD. AND D. WESTERN THERAPEUTICS INSTITUTE, INC.**
Fecha de Solicitud : 29 de mayo de 2007

1. Yo, Luz Clemencia DE PÁEZ, en mi condición de apoderada de la sociedad solicitante de la patente de la referencia, de acuerdo con la sustitución del poder a mí otorgada por el doctor Emilio Ferrero, me refiero al oficio No. **14794** notificado por fijación en lista del 12 de noviembre de 2010.

2. Mediante oficio No. **14794** se puso en conocimiento del solicitante el informe que contiene la solicitud hecha por su Despacho para la presente solicitud de patente. Atiendo a las observaciones de la manera siguiente:

2.1 Presentación de documentos solicitados

Mediante oficio No. **14794** y de acuerdo con las disposiciones del Artículo 46 de la Decisión 486 su Despacho solicita el siguiente documento en inglés:

2.1.1 WO 99/20620 – Isoquinoline derivatives and drug

Con el fin de atender a dicha solicitud, se presenta junto con este memorial copia del documento mencionado.

3. Puesto que se han atendido a cabalidad las observaciones hechas por su Despacho sobre la presente solicitud de patente, atentamente solicito que se continúe su tramitación.

4. ANEXOS

- Copia en inglés del documento WO 99/20620 – Isoquinoline derivatives and drug.
- Copia auténtica de la sustitución de poder efectuada a mi favor por el Doctor Emilio FERRERO.

Señora Directora,

Luiz Clemencia Sobríz
LUZ CLEMENCIA DE PÁEZ

CC. 35.456.344 de Usaquén

T.P.A. No. 23.555

 COLO-15943-841-001-5
DVF\DV\FID-186618\WPC15943
No. M-2011-186618\DV F

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Señores

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO

E. S. D.

Poderdante : KOWA CO., LTD.

Poder conferido el : 26/07/2005

Yo, EMILIO FERRERO, obrando como apoderado principal del poderdante arriba mencionado, mediante el presente escrito sustituyo el poder a mi otorgado a la doctora LUZ CLEMENCIA DE PAEZ, identificada con cédula de ciudadanía No. 35.456.344 y Tarjeta Profesional N°23.555, con todas las facultades que me fueron conferidas.

Hago la anterior sustitución conforme al artículo 66 del Código de Procedimiento Civil.

Señora Jefe,

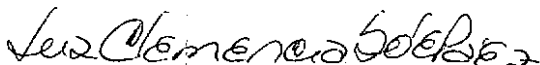


EMILIO FERRERO

T.P.A. N° 31.929

C.C. N° 438.019 de Usaquén

Acepto:


LUZ CLEMENCIA DE PAEZ

T.P.A. N° 23.555

C.C. No. 35.456.344 de Usaquén



2

NOTARIA SEXTA DE BOGOTÁ
RECONOCIMIENTO Y PRESENTACION PERSONAL
Bogotá, D.C. **3 - NOV. 2009**
Ante mí AMPARO QUINTERO ARTURO,
NOTARIA SEXTA DEL CÍRCULO DE
BOGOTÁ, D.C.
Comparecido(s) *Emilio Feryero*
Williamson Loz Clemente
Suarez de Paez
Quien(s) exhibi(aron) la(s) C.C.
438019 T.P. 31923
35456344 T.P. 23555
Y declaró(aron) que la(s) firma(s) que aparece(n)
en el presente documento es(son) la(s) suya(s) y
que el contenido del mismo es cierto.
En constancia se firma esta diligencia.

Emilio Feryero
Luz Clemencia Suarez de Paez



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Señores

SUPERINTENDENCIA DE INDUSTRIA Y COMERCIO

E. S. D.

Poderdante : D. **WESTERN**
THERAPEUTICS INSTITUTE, INC.

Poder conferido el : 14/05/2007

Yo, EMILIO FERRERO, obrando como apoderado principal del poderdante arriba mencionado, mediante el presente escrito sustituyo el poder a mi otorgado a la doctora LUZ CLEMENCIA DE PAEZ, identificada con cédula de ciudadanía No. 35.456.344 y Tarjeta Profesional N°23.555, con todas las facultades que me fueron conferidas.

Hago la anterior sustitución conforme al artículo 66 del Código de Procedimiento Civil.

Señora Jefe,



EMILIO FERRERO

T.P.A. N° 31.929

C.C. N° 438.019 de Usaquén

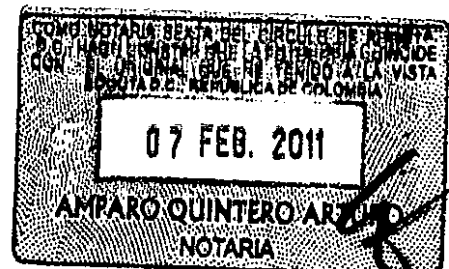
Acepto:



LUZ CLEMENCIA DE PAEZ

T.P.A. N° 23.555

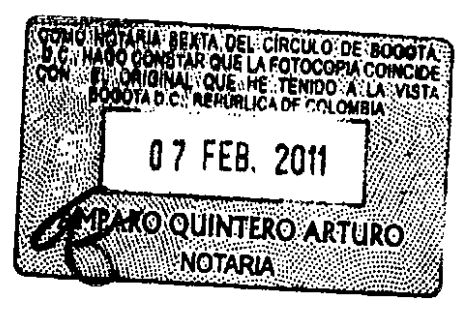
C.C. No. 35.456.344 de Usaquén



4

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Bogotá, D.C. **3 - NOV. 2009**
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NOTARIA SEXTA DEL CÍRCULO DE
BOGOTÁ, D.C.
Comparación(aron) *Emilio Ferrero*
Williamson *duz clemencia*
Suarez de paez
Quien(s) exhibió(aron) la(s) C.C.
438019 TP 31929
35456344 TP 28555
Y declaró(aron) que la(s) firma(s) que aparece(n)
en el presente documento es(son) la(s) suya(s) y
que el contenido del mismo es cierto.
En constancia se firma esta diligencia.

Emilio Ferrero
Suarez Clemencia Suarez



ISOQUINOLINE DERIVATIVE AND MEDICINE

TECHNICAL FIELD

The present invention relates to an isoquinoline derivative which has cerebral vasospasm-inhibitory activity and blood flow-increasing activity and is of value as a medicine.

BACKGROUND ART

Cerebrovascular diseases and heart diseases are nowadays counted as major causes of death in many developed countries, like cancer is so.

Cerebrovascular diseases are roughly classified into a hemorrhagic group and an ischemic group. Representative of the ischemic group are subarachnoid hemorrhage arising from brain aneurismal rupture, hypertensive cerebral hemorrhage, and head trauma. Subarachnoid hemorrhage entails a luminal constriction of the trunk arteries of the brain beginning 4 to 5 days after the bleeding episode and persisting for about 2 weeks. This is the condition known as cerebral vasospasm. Much remains to be known as yet about its mechanism of onset but a variety of physiologically active substances (such as catecholamines, hemoglobin, peroxy lipids, arachidonic acid metabolites, peptides, protein kinase C, etc.) are suspected to be involved and their

interactions are considered to be etiologic factors. As the vascular stenosis and consequent ischemia give rise to neurological disorders, not only the functional prognosis but sometimes the vital prognosis is critically affected. The ischemic group is represented by cerebral infarction and transient ischemic attack (TIA). The vascular lesion and neuronal damage caused by ischemia or hemorrhage lead to numbness, dyskinesia such as motor paralysis of the limbs, and neuropsychiatric disorders in the acute through chronic stages and even to death through disturbance of consciousness in severe cases.

In the treatment of those cerebrovascular diseases, antithrombotics and nootropic drugs are used. However, few drugs are available which may inhibit life-threatening cerebral vasospasms and neuronal damages progressive to dementia and thus, there has been a standing demand for effective therapeutic drugs. As a therapeutic drug for the cerebral vasospasm secondary to subarachnoid hemorrhage, fasudil hydrochloride [hexahydro-1-(5-isoquinolinylsulfonyl)-1H-1,4-diazepine] is the only remedy in clinical use today [cf. Basic Pharmacology & Therapeutics, 1992, 20 (suppl. 6), 1515, among others] but this compound is not a fully satisfactory drug from the standpoint of efficacy and side effect.

On the other hand, angina pectoris is a typical ischemic heart disease. In angina pectoris, ischemia caused by vascular stenosis induces myocardial infarction which may lead to death. The current pharmacotherapy is performed primarily with vasodilators, but many of the vasodilators simultaneously have a hypotensive activity. Moreover, few are effective enough to control vasospasms.

It is known that compounds having an isoquinoline nucleus substituted by a cycloaminosulfonyl group in the 5-position are of value as circulatory system-improving drugs (vasodilators, nootropic drugs, antianginal drugs, prophylactic and therapeutic drugs for thrombosis in the cerebrovascular and cardiovascular systems, antihypertensives, and brain function improving agents) (Japanese Kokai Tokkyo Koho (briefly KTK) S57-156463, KTK S57-200366, KTK S58-121278, KTK S58-121279, KTK S59-93054, KTK S60-81168, KTK S61-152658, and KTK S61-227581). Furthermore, those compounds are known to be useful as cerebral function-improving drugs [drugs for prevention, amelioration or retardation of progression of disturbances of the brain tissue function and status (inclusive of metabolism) and the associated symptoms and sequelae] (Kokai Tokkyo Koho H2-256617).

The inventors of the present invention discovered

that compounds containing an isoquinoline nucleus substituted by a cycloaminosulfonyl group in the 5-position and having a substituent in the 4-position are superior to said known compounds as prophylactic or therapeutic drugs for cerebrovascular diseases. And the inventors previously filed a patent application claiming said compounds (see WO 97/28130). The compound of the present invention which has fluorine in the 4-position and a hexahydro-4H-1,4-diazepin-1-ylsulfonyl group in the 5-position of an isoquinoline nucleus and in which the 2-position of said hexahydro-4H-1,4-diazepine moiety has been substituted by methyl has not been specifically disclosed in the above application.

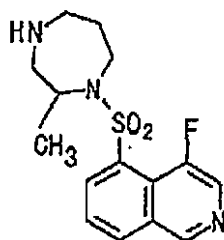
DISCLOSURE OF THE INVENTION

The object of the present invention is to provide a compound which is highly safe and superior to any known compound as a prophylactic and/or therapeutic drug for cerebrovascular diseases, particularly as a cerebral vasospasm inhibitor and a prophylactic and/or therapeutic drug for diseases accompanied by ischemic lesions.

To accomplish the above object, the inventors of the present invention synthesized a large number of structurally novel compounds and discovered, in the

course of their screening, that a compound of the following formula [I] shows cerebral vasospasm-inhibitory activity at a very low dose level and, in addition, exhibits an excellent blood flow-increasing activity by dilating the coronary and cerebral blood vessels without lowering the systemic blood pressure.

The present invention, therefore, is directed to a pharmaceutical composition comprising a compound of the following formula [I] or a pharmaceutically acceptable salt thereof as an active ingredient.



[I]

In terms of chemical structure, the compound of the invention is characterized in that the 4-position of its isoquinoline nucleus has been substituted by fluorine and that the 2-position of the hexahydro-4H-1,4-diazepine ring of the hexahydro-4H-1,4-diazepin-1-ylsulfonyl group in the 5-position of said nucleus has been substituted by methyl.

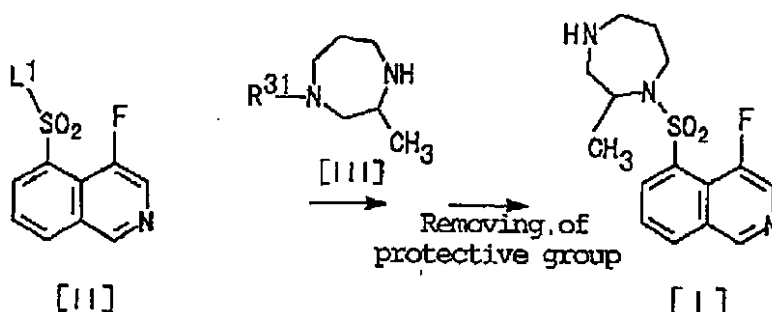
The compound [I] according to the invention has asymmetric carbon and, as such, may occur as optical

isomers. Those respective isomers as well as mixtures thereof also fall within the scope of the invention. Among them, the (S) compound is preferred.

The "pharmaceutically acceptable salt" includes salts with inorganic acids such as hydrochloric acid, sulfuric acid, nitric acid, phosphoric acid, hydrofluoric acid, hydrobromic acid, etc. and salts with organic acids such as acetic acid, tartaric acid, lactic acid, citric acid, fumaric acid, maleic acid, succinic acid, methanesulfonic acid, ethanesulfonic acid, benzenesulfonic acid, toluenesulfonic acid, naphthalenesulfonic acid, camphorsulfonic acid, etc.

The compound [I] according to the invention can be produced by the process disclosed in WO 97/28130 or the processes described below.

Process 1



(wherein R^{31} represents a protective group; L^1 represents hydroxy or a leaving group)

The leaving group L^1 includes the residue of a

reactive derivative of sulfonic acid which is described hereinafter. The protective group R^{31} includes acyl groups such as formyl, acetyl, benzoyl, etc., aralkyloxycarbonyl groups such as benzyloxycarbonyl etc., alkoxycarbonyl groups such as tert-butoxycarbonyl etc., and aralkyl groups such as benzyl.

The amine of the formula [III] is reacted with a sulfonic acid of the formula [II] or a reactive derivative in a suitable solvent and the protective group is then removed to give compound [I]. The reaction solvent may be any solvent that does not interfere with the reaction and includes but is not limited to ethers such as tetrahydrofuran (THF), dioxane, diethyl ether, etc.; hydrocarbons such as benzene, toluene, etc.; halogenated hydrocarbons such as methylene chloride, chloroform, etc.; aprotic solvents such as N,N-dimethylformamide, N,N-dimethylacetamide, etc.; pyridine; acetonitrile; etc. and mixtures thereof. The reactive derivative of sulfonic acid includes sulfonyl halides (e.g. sulfonyl chloride, sulfonyl bromide, etc.), sulfonic anhydride, N-sulfonylimidazolidine, etc. Particularly preferred is a sulfonyl halide.

This reaction is preferably carried out in the presence of a suitable base. The base includes inorganic bases such as alkali metal hydrogencarbonates (e.g.

sodium hydrogencarbonate), alkali metal carbonates (e.g. potassium carbonate), and alkali metal hydroxides (e.g. sodium hydroxide, potassium hydroxide) and organic tertiary amines such as triethylamine, triethylenediamine, etc. Use of a basic solvent such as pyridine as the reaction solvent is advantageous in that the use of said base could be omitted.

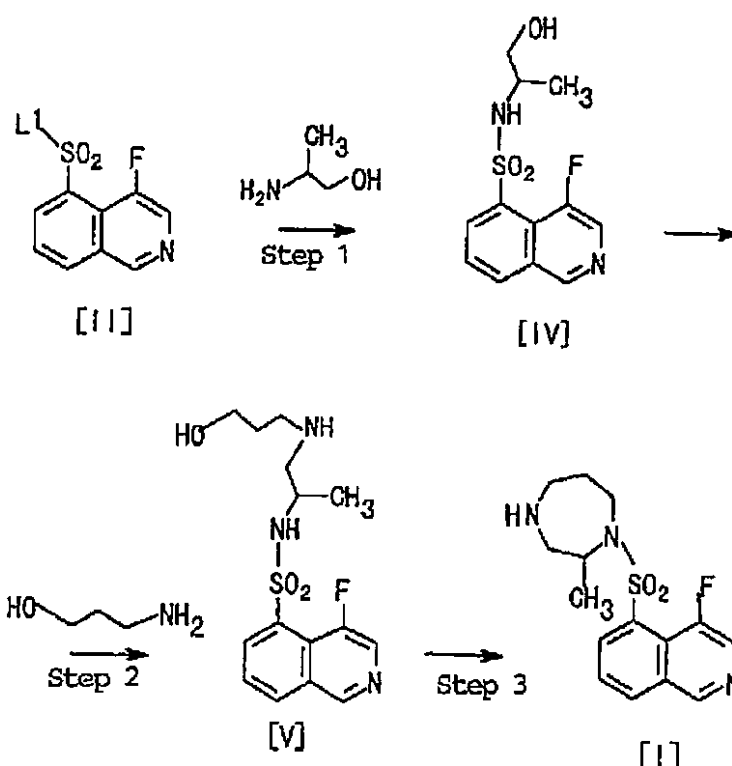
Usually this reaction proceeds at room temperature but, where necessary, can be conducted under cooling or heating within a temperature range of -78 to 150°C , preferably 0 to 120°C . When the base is used, the amount of said amine [III] is preferably 1 to 5 molar equivalents, based on said reactive derivative [II], and more preferably 1 to 2 molar equivalents on the same basis. The amount of the base is preferably 1 to 10 molar equivalents and more preferably 1 to 3 molar equivalents, based on said amine [III]. When the base is not used, the amount of sulfonic acid or its reactive derivative [II] is preferably equimolar or less, more preferably in the range of 0.5 to 0.1 molar equivalent, based on amine [III]. The reaction time, which depends on the species of starting compounds, the kind of solvent, the reaction temperature, etc., is generally 5 minutes to 70 hours. The protective group is then removed by a per se known procedure. Thus, the protective group may be

removed by such known procedures as acid treatment, alkali treatment, catalytic reduction, etc.

The starting compound [II] can be produced in the same manner as Reference Example 1 which is given hereinafter.

The starting compound [III] can be produced in the same manner as Reference Example 2.

Process 2



(wherein R³¹ and L¹ are as defined hereinbefore)

(Step 1) 2-Amino-1-propanol is reacted with compound [II] in the same manner as described in Process 1 to give compound [IV].

(Step 2) The hydroxyl group of compound [IV] is

converted to halogen (e.g. chlorine, bromine), sulfonyloxy, or acyloxy (e.g. tosyloxy, methane-sulfonyloxy, acetyloxy) and, then, 3-amino-1-propanol is permitted to react in a suitable solvent either in the presence or the absence of a base in the same manner as described in Process 1 to give compound [V].

(Step 3) The secondary amino nitrogen atom of compound [V] is protected by a per se known procedure and, then, the compound is cyclized in a suitable solvent in the presence of a base, followed by deprotection to give compound [I].

Compound [I] can also be produced by treating compound [V] with triphenylphosphine and a dialkyl azodicarboxylate to cause an intramolecular dehydrative condensation.

In the above process, where necessary the amino group is protected with a conventional protective group beforehand and after completion of the reaction, the protective group is removed by a per se known procedure such as acid treatment, alkali treatment, or catalytic reduction. The amino-protecting group which can be used includes but is not limited to benzyl, benzyloxycarbonyl, and trifluoroacetyl.

The compound [I] of the invention is usually obtained as a racemic compound. The racemic compound

as such has pharmacological activity but, if desired, can be resolved into the respective isomers. For example, this optical resolution can be achieved by a known method, for example by reacting the compound with an optically active carboxylic acid (e.g. (+)- or (-)-tartaric acid, (+)- or (-)-malic acid) or an optically active sulfonic acid compound (e.g. (+)-camphorsulfonic acid) to give a salt and carrying out fractional recrystallization or by the method using a chiral column. The optical isomers can also be respectively provided by conducting the reaction using a chiral compound (S-configured or R-configured) for starting compound [III].

The compound [I] according to the invention can be converted to said salt by the known technology. For example, the hydrochloride of compound [I] according to the invention can be provided by dissolving compound [I] in a solution of hydrogen chloride in an alcohol or ethyl ether, concentrating the solution, and harvesting the resulting crystals.

The compound of the invention, thus produced, can be isolated and purified in the form of a free base or an acid addition salt by per se known procedures such as concentration, pH change, redistribution, solvent extraction, crystallization, fractional distillation, chromatography, and recrystallization.

The compound of the invention has cerebral vasospasm-inhibitory activity and is highly safe so that it is useful for the prevention and treatment of cerebrovascular diseases, particularly organic brain damages due to cerebral vasospasms secondary to cerebral hemorrhage. Moreover, because it has blood flow-increasing activity, the compound is useful as a prophylactic or therapeutic drug for lesions associated with ischemic diseases.

When the compound of the invention is to be administered as a medicine, the compound is administered to a mammal inclusive of man, either as it is or in the form of a pharmaceutical composition containing 0.1% to 99.5%, preferably 0.5% to 90%, of the compound in a pharmaceutically acceptable, nontoxic and inert carrier.

As the carrier, at least one kind of solid, semisolid, or liquid diluent, filler and other auxiliary formulating agents is used. The pharmaceutical composition is preferably administered in unit dosage forms. The pharmaceutical composition of the invention can be administered orally, parenterally (into tissues), topically (e.g. transdermally), or rectally. Of course, a dosage form suited to each route of administration should be chosen. Particularly preferred is

intravenous administration or oral administration.

The dosage as a drug is preferably adjusted in consideration of patient factors, such as age and body weight, the route of administration, and the nature and severity of illness but when it is used as a prophylactic and/or therapeutic drug for cerebral vasospasm, the recommended intravenous dosage for an adult human per day is 0.1 to 100 mg/man, preferably 1 to 30 mg/man, as the active ingredient. In the case of oral administration, the daily dose may range from 1 to 1,000 mg/man, preferably 10 to 30 mg/man. When the composition is to be administered intravenously as a prophylactic and/or therapeutic drug for diseases accompanied by ischemic lesions, the active ingredient is used in the daily dose range of 0.1 to 100 mg/man, preferably 1 to 30 mg/man. When the oral route is selected, the daily dose range is 1 to 1,000 mg/man, preferably 10 to 30 mg/man. A higher dosage may be justified in some cases, while a lower dosage may suffice in other cases. The above-mentioned daily dosage can be administered in a few divided doses.

BEST MODE FOR CARRYING OUT THE INVENTION

The present invention is now described in further detail by way of reference examples relating to the production of representative starting compounds,

examples of production of the compound of the invention, formulation examples for representative species of the compound, and test examples using the compound. Those examples, however, should not be construed as defining the scope of the invention. Incidentally, the optical rotation data were generated at 20°C.

Reference Example 1

Synthesis of 5-chlorosulfonyl-4-fluoroisoquinoline

(Step 1)

A 300 ml sealed tube was charged with 160 ml of 29% aqueous ammonia, 3 g of copper sulfate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$), and 49.92 g of 4-bromoisoquinoline and the charge was stirred at 170°C for 16 hours. After cooling, the reaction mixture was diluted with 40 ml of 2N sodium hydroxide solution and extracted with 400 ml of benzene for a total of 5 times. The organic layer was dehydrated over anhydrous magnesium sulfate and treated with activated carbon and the solvent was then distilled off to provide 29.5 g of 4-aminoisoquinoline.

(Step 2)

In 100 ml of 42% hydrofluoric acid was dissolved 28.8 g of 4-aminoisoquinoline. This solution was cooled with dry ice-ether and as it began to become solidified, 13.8 g of sodium nitrite was added over 1 hour. This reaction mixture was filtered and the resulting solid

(diazonium salt) was quickly washed with ether at low temperature to remove as much water as possible. This diazonium salt was poured in xylene and heated with a hot air current (in a dryer) under stirring. After gases began to evolve, the mixture was stirred at room temperature for 2 hours. The xylene was then separated from the tarry product by decantation and the xylenic solution was extracted with 100 ml of diluted hydrochloric acid 3 times. The HCl solution layer and the tarry residue were combined, diluted with 100 ml of water, made basic with potassium carbonate and extracted with 400 ml of ether 3 times. The extract was dehydrated over anhydrous magnesium sulfate and filtered. Active charcoal was added to the filtrate and the mixture was filtered with the aid of Celite. The solvent was then distilled off and the residue was purified by silica gel column chromatography (chloroform). Finally the solvent was distilled off to recover 14.5 g of 4-fluoroisoquinoline.

(Step 3)

To 90 ml of sulfuric acid was added 22.05 g of 4-fluoroisoquinoline. To this mixture under ice cooling was added a solution of potassium nitrate (18.18 g) in sulfuric acid (90 ml) dropwise, and the mixture was stirred at room temperature for 2 hours. This

reaction mixture was poured onto ice and made basic with aqueous ammonia under cooling. The resulting crystals were harvested, rinsed with water, and dried to provide 22.16 g of 4-fluoro-5-nitroisoquinoline.

(Step 4)

In 60 ml of ethanol was suspended 23.04 g of 4-fluoro-5-nitroisoquinoline. To this suspension was added 144 ml of concentrated hydrochloric acid, and under ice-cooling and stirring, a solution of stannous chloride dihydrate (90.24 g) in ethanol (144 ml) was added dropwise. The mixture was then stirred at room temperature for 3 hours. This reaction mixture was concentrated, diluted with 200 ml of iced water, made basic with 2N sodium hydroxide, and extracted with 500 ml of chloroform 3 times. The chloroform layer was dehydrated over anhydrous magnesium sulfate and concentrated and the residue was purified by silica gel column chromatography (chloroform:methanol = 100:1) to provide 15.05 g of 5-amino-4-fluoroisoquinoline.

(Step 5)

In 100 ml of concentrated hydrochloric acid was suspended 8.1 g of 5-amino-4-fluoroisoquinoline. To this suspension was added 5.18 g of sodium nitrite in 20 ml of water dropwise at a temperature not over -5°C , and the mixture was stirred at room temperature

for 30 minutes (preparation of Solution A). Separately, a solution of cupric chloride dihydrate (2.83 g) in water (12.5 ml) was added to 62.5 ml of acetic acid saturated with sulfurous acid gas and the mixed solution was added dropwise to Solution A. This reaction mixture was further stirred at room temperature for 1 hour and then incubated on a water bath at 30°C to remove the unreacted sulfurous acid. The reaction mixture was extracted with 400 ml of chloroform 3 times and the extract was neutralized and washed with aqueous sodium hydrogencarbonate solution and dehydrated. The solvent was finally distilled off to provide 9.6 g of 5-chlorosulfonyl-4-fluoroisoquinoline.

Reference Example 2

Synthesis of (S)-hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine
(Step 1)

In 150 ml of chloroform was dissolved 15 g of (S)-(+)-2-amino-1-propanol, followed by addition of 20.21 g of triethylamine. To this mixture was added a solution of benzyloxycarbonyl chloride (ZCl) (34.07 g) in chloroform (50 ml) gradually dropwise under ice-cooling and stirring, and the mixture was further stirred at room temperature overnight. This reaction mixture was diluted with 200 ml of water and extracted

with 200 ml of chloroform 3 times. The organic layer was washed with 200 ml of saturated aqueous sodium chloride solution twice and dehydrated over anhydrous magnesium sulfate and the solvent was distilled off. The crystalline residue was triturated in 200 ml of n-hexane and the resulting powders were recovered by filtration, rinsed with n-hexane, and dried to provide 37.04 g of (S)-2-benzyloxycarbonylamino-1-propanol. (Step 2)

In 140 ml of methylene chloride was dissolved 36.52 g of (S)-2-benzyloxycarbonylamino-1-propanol, followed by addition of 18.67 g of triethylamine. To this mixture was added a solution of mesyl chloride (21.40 g) in methylene chloride (60 ml) gradually dropwise under ice-cooling and stirring, and the mixture was stirred for 20 minutes. This reaction mixture was diluted with 200 ml of water and extracted with 200 ml of chloroform 3 times. The organic layer was washed with 200 ml of saturated aqueous sodium chloride solution and dehydrated over anhydrous magnesium sulfate and the solvent was distilled off to recover 49.53 g of (S)-2-benzyloxycarbonylamino-1-methanesulfonyloxypropane.

(Step 3)

In 60 ml of tetrahydrofuran (THF) was dissolved

70.0 g of 3-amino-1-propanol. To this solution was added a suspension of (S)-2-benzyloxycarbonylamino-1-methanesulfonyloxypropane (48.97 g) in THF (140 ml) at room temperature, and the mixture was refluxed at 80°C with stirring overnight. After cooling, the reaction mixture was diluted with 200 ml of water and extracted with 200 ml of chloroform 3 times. The organic layer was washed with 200 ml of saturated aqueous sodium chloride solution twice and dehydrated over anhydrous magnesium sulfate and the solvent was distilled off to provide 37.61 g of (S)-6-benzyloxycarbonylamino-4-azaheptan-1-ol as light-yellow oil. This oil crystallized as it was left standing awhile.

(Step 4)

In 150 ml of THF was dissolved 37.1 g of (S)-6-benzyloxycarbonylamino-4-azaheptan-1-ol. To this solution was added a solution of di-tert-butyl carbonate (34.7 g) in THF (50 ml) dropwise under ice-cooling and stirring, and the mixture was further stirred at room temperature overnight. This reaction mixture was diluted with 200 ml of water and extracted with 200 ml of chloroform 3 times. The organic layer was washed with 200 ml of saturated aqueous sodium chloride solution twice and dehydrated over anhydrous

magnesium sulfate and the solvent was distilled off. The residue was purified by silica gel column chromatography (Wako-gel C-200/TM, 1 kg; chloroform: methanol = 100:1→100:5) to provide 44.42 g of (S)-6-benzyloxycarbonylamino-4-(tert-butoxycarbonyl)-4-azaheptan-1-ol as light-yellow oil.

(Step 5)

In 50 ml of methylene chloride was dissolved 10.0 g of

(S)-6-benzyloxycarbonylamino-4-(tert-butoxycarbonyl)-4-azaheptan-1-ol, followed by addition of 3.0 g of triethylamine, and the mixture was stirred under ice-cooling. To this mixture was added a solution of mesyl chloride (3.32 g) in methylene chloride (25 ml), and the mixture was stirred at room temperature for 10 minutes. This reaction mixture was diluted with 200 ml of water and extracted with 100 ml of chloroform 3 times. The organic layer was washed with 200 ml of saturated aqueous sodium chloride solution and dehydrated over anhydrous magnesium sulfate and the solvent was distilled off to recover 11.74 g of

(S)-6-benzyloxycarbonylamino-4-(tert-butoxycarbonyl)-4-azaheptyl methanesulfonate as light-yellow oil.

(Step 6)

In dimethyl sulfoxide (DMSO) was dissolved 11.74

g of (S)-6-benzyloxycarbonylamino-4-tert-butoxycarbonyl)-4-azaheptyl methanesulfonate, followed by addition of 2.2 g of 60% sodium hydride, and the mixture was stirred at room temperature for 30 minutes. This reaction mixture was diluted with water and ethyl acetate and the organic layer was washed with water and then with 200 ml of saturated aqueous sodium chloride solution 5 times and dehydrated over anhydrous magnesium sulfate. The solvent was then distilled off and the residue was purified by silica gel column chromatography (Wako-gel C-200, TM, 200 g; eluent: chloroform) to provide 8.53 g of (S)-hexahydro-1-benzyloxycarbonyl-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine as light-yellow oil.
(Step 7)

Using an atmospheric pressure reduction apparatus of 500 ml capacity, 8.53 g of (S)-hexahydro-1-benzyloxycarbonyl-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine was dissolved in 80 ml of methanol and after addition of 4.0 g of 5% palladium-on-carbon, catalytic reduction under atmospheric pressure was carried out at room temperature. After 6 hours, the reaction mixture was filtered with the aid of Celite and the solvent was distilled off to provide 3.97 g of (S)-hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-

1,4-diazepine as light-yellow oil.

In the same manner as above, (R)-hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine and hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine were synthesized.

Example 1

Hexahydro-1-(4-fluoro-5-isoquinolinesulfonyl)-
2-methyl-1H-1,4-diazepine

(1) To a mixed solution of hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine (0.51 g) and triethylamine (0.30 g) in methylene chloride (10 ml) was added 0.49 g of 5-chlorosulfonyl-4-

fluoroisoquinoline, and the mixture was stirred at room temperature for 12 hours. This reaction mixture was concentrated and the residue was dissolved in methylene chloride. This methylene chloride solution was washed with water, dehydrated over anhydrous magnesium sulfate, and concentrated. The residue was purified by silica gel column chromatography (chloroform/methanol = 100/1) to provide 0.51 g of hexahydro-4-(tert-butoxycarbonyl)-1-(4-fluoro-5-isoquinolinesulfonyl)-2-methyl-1H-1,4-diazepine.

(2) In 10 ml of methylene chloride was dissolved 0.99 g of the compound obtained in (1), and 10 ml of trifluoroacetic acid was added dropwise. The mixture

was stirred at room temperature for 2 hours. This reaction mixture was neutralized with saturated aqueous sodium hydrogencarbonate solution and extracted with chloroform. The chloroform layer was dehydrated over anhydrous magnesium sulfate and concentrated. The residue was purified by silica gel column chromatography (chloroform/methanol = 10/1) to provide 0.24 g of the objective compound.

NMR spectrum (CDCl_3): δ (ppm) 0.91 (3H, d), 1.69-1.88 (3H, m), 2.47-2.77 (2H, m), 3.13-3.36 (3H, m), 3.92-4.10 (2H, m), 7.73 (1H, t), 8.22 (1H, dd), 8.57 (1H, d), 8.84 (1H, d), 9.15 (1H, s).

Example 2

(S)-(-)-Hexahydro-1-(4-fluoro-5-isoquinoline-sulfonyl)-2-methyl-1H-1,4-diazepine hydrochloride

(1) To a mixed solution of (S)-hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine (0.54 g) and triethylamine (0.38 g) in methylene chloride (10 ml) was added 0.62 g of 5-chlorosulfonyl-4-fluoro-isoquinoline, and the mixture was stirred at room temperature for 12 hours. This reaction mixture was concentrated and the residue was dissolved in methylene chloride. The methylene chloride solution was washed with water, dehydrated over anhydrous magnesium sulfate, and concentrated. The residue was

purified by silica gel column chromatography (chloroform/methanol = 100/1) to provide 0.99 g of (S)-hexahydro-4-(tert-butoxycarbonyl)-1-(4-fluoro-5-isoquinolinesulfonyl)-2-methyl-1H-1,4-diazepine.

(2) In 10 ml of methylene chloride was dissolved 0.99 g of the compound obtained in (1), followed by dropwise addition of trifluoroacetic acid, and the mixture was stirred at room temperature for 2 hours. This reaction mixture was neutralized with saturated aqueous sodium hydrogencarbonate solution and extracted with chloroform. The chloroform layer was dehydrated over anhydrous magnesium sulfate and concentrated. The residue was purified by silica gel column chromatography (chloroform/methanol = 10/1) to provide 0.58 g of (S)-hexahydro-1-(4-fluoro-5-isoquinolinesulfonyl)-2-methyl-1H-1,4-diazepine.

NMR spectrum (CDCl_3): δ (ppm) 0.91 (3H, d), 1.69-1.88 (3H, m), 2.47-2.77 (2H, m), 3.13-3.36 (3H, m), 3.92-4.10 (2H, m), 7.73 (1H, t), 8.22 (1H, dd), 8.57 (1H, d), 8.84 (1H, d), 9.15 (1H, s).

(3) In 10 ml of methanol was dissolved 0.58 g of the compound obtained in (2), followed by addition of 1.80 ml of 1N-hydrochloric acid, and the mixture was concentrated to provide 0.50 g of the objective compound.

Elemental analysis ($\text{C}_{15}\text{H}_{18}\text{FN}_3\text{O}_2\text{S HCl}$)

Calcd. (%) C, 50.01; H, 5.32; N, 11.68

Found (%) C, 50.26; H, 5.60; N, 11.44

$[\alpha]_D = -9.02^\circ$ (c=1.086, H₂O)

Example 3

(R) - (+) - Hexahydro-1-(4-fluoro-5-iso-quinolinesulfonyl)-2-methyl-1H-1,4-diazepine hydrochloride

(1) Using 0.54 g of (R)-hexahydro-4-(tert-butoxycarbonyl)-2-methyl-1H-1,4-diazepine, the procedure of Example 2 (1) and (2) was otherwise repeated to provide (R) - (+) - hexahydro-1-(4-fluoro-5-iso-quinolinesulfonyl)-2-methyl-1H-1,4-diazepine.

NMR spectrum (CDCl₃): δ (ppm) 0.91 (3H, d), 1.69-1.88 (3H, m), 2.47-2.77 (2H, m), 3.13-3.36 (3H, m), 3.92-4.10 (2H, m), 7.73 (1H, t), 8.22 (1H, dd), 8.57 (1H, d), 8.84 (1H, d), 9.15 (1H, s).

(2) Using the compound obtained in (1) above, the procedure of Example 2 (3) was otherwise repeated to provide 0.45 g of the objective compound.

Elemental analysis (C₁₅H₁₈FN₃O₂S HCl)

Calcd. (%) C, 50.01; H, 5.32; N, 11.68

Found (%) C, 50.27; H, 5.05; N, 11.82

$[\alpha]_D = 7.70^\circ$ (c=1.116, H₂O)

Example 4

(S) - (-) - Hexahydro-1-(4-fluoro-5-iso-

quinolinesulfonyl)-2-methyl-1H-1,4-diazepine
hydrochloride (an alternative process)

(1) In 100 ml of methylene chloride were dissolved 3.00 g of (S)-(+)-2-amino-1-propanol and 6.06 g of triethylamine, followed by addition of 9.82 g of 5-chlorosulfonyl-4-fluoroisoquinoline, and the mixture was stirred at room temperature for 12 hours. This reaction mixture was washed with water, dehydrated over anhydrous magnesium sulfate, and concentrated. The resulting crystal crop was washed with isopropyl ether to provide 7.96 g of (S)-4-fluoro-5-[N-(1-hydroxypropan-2-yl)aminosulfonyl]isoquinoline.

(2) In 30 ml of methylene chloride were dissolved 1.42 g of the compound obtained in (1) and 0.76 (g) of triethylamine, and 0.63 g of methanesulfonyl chloride was added dropwise under ice-cooling. The mixture was then stirred at room temperature for 1 hour. This reaction mixture was washed with water, dehydrated over anhydrous magnesium sulfate, and concentrated. To the residue was added 20 ml of THF, and 1.88 g of 3-aminopropanol was added dropwise. The mixture was then stirred at room temperature for 12 hours. This reaction mixture was concentrated and the residue was purified by silica gel column chromatography (methylene chloride/methanol/29% aqueous ammonia = 90/10/1) to

provide 1.56 g of (S)-4-fluoro-5-[N-(1-hydroxy-4-azaheptan-6-yl)aminosulfonyl]isoquinoline.

(3) In 15 ml of THF were dissolved 1.54 g of the compound obtained in (2) and 1.77 g of triphenylphosphine, and 3.41 g of 40% diisopropyl ester of azodicarboxylic acid/toluene was added dropwise. The mixture was then stirred at room temperature for 2 hours. This reaction mixture was concentrated and the residue was diluted with ethyl acetate and extracted with 1N-hydrochloric acid. The aqueous layer was made weakly basic with sodium hydrogencarbonate and the resulting oil was extracted with chloroform. The chloroform layer was washed with water, dehydrated over anhydrous magnesium sulfate, and concentrated. The residue was dissolved in ethanol and 1N-hydrogen chloride/ether was added. The resulting crystals were harvested by filtration and rinsed with ethanol to provide 0.60 g of the objective compound.

Elemental analysis ($C_{15}H_{19}FN_3O_2S \cdot HCl$)

Calcd. (%) C, 50.01; H, 5.32; N, 11.68

Found (%) C, 49.82; H, 5.21; N, 11.43

m.p. 258-260°C

Formulation Example 1

Recipe (in each ml)

Compound of Example 2	3 mg
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Sodium chloride	9 mg
Water for injection	q.s.
	1 ml

Preparation method:

The compound of Example 2 and sodium chloride in the above ratio are dissolved in water for injection and the solution is filtered through a membrane filter (0.22 μ m), filled into ampoules, and sterilized to provide an aqueous injectable product.

Formulation Example 2

Recipe (in each vial)

Compound of Example 2	3 mg
Mannitol	50 mg

Preparation Method:

The compound of Example 2 and mannitol in the above ratio are dissolved in water for injection, filtered aseptically through a membrane filter (0.22 μ m), and the filtrate is filled into vials and lyophilized in the routine manner to provide an injectable product of the extemporaneous reconstitution type.

Formulation Example 3

Recipe (in each 180 mg tablet)

Compound of Example 2	10 mg
Lactose	100 mg
Corn starch	55 mg

Low-substitution-degree

hydroxypropylcellulose	9 mg
Polyvinyl alcohol	
(partially hydrolyzed)	5 mg
Magnesium stearate	1 mg

Preparation method:

The above components except for polyvinyl alcohol and magnesium stearate are evenly blended in the above ratio and using an aqueous solution of polyvinyl alcohol as a binder, a granulation for compression is prepared by the wet granulation method. Magnesium stearate is mixed with the above granulation and the resulting composition is compressed with a tablet machine to provide oral tablets each weighing 180 mg.

Formulation Example 4

Recipe (in each 220 mg capsule)

Compound of Example 2	10 mg
Lactose	187 mg
Microcrystalline cellulose	20 mg
Magnesium stearate	3 mg
	220 mg

Preparation method:

The components in the above ratio are evenly blended and using a capsule filling machine, the composition (220 mg) is filled into hard capsule shells to provide

a hard capsule product.

Formulation Example 5

Recipe (in each g of granules)

Compound of Example 2	10 mg
Lactose	880 mg
Low-substitution-degree hydroxypropylcellulose	70 mg
Hydroxypropylcellulose	40 mg
	1000 mg

Preparation method:

The components other than hydroxypropylcellulose in the above ratio are evenly blended and using an aqueous solution of hydroxypropylcellulose as a binder, the mixture is kneaded and mechanically granulated to provide granules.

The following are test examples for a representative compound of the invention. The compound of Example 2 was used as the investigational substance. As reference compounds, hexahydro-1-(4-fluoro-5-isoquinolinesulfonyl)-1H-1,4-diazepine hydrochloride (hereinafter referred to as Compound A), which is specifically disclosed in WO 97/28130 and resembling the compound of the invention most closely in chemical structure, and the commercial product fasudil hydrochloride were used.

Test Example 1

Action on the calcium ionophore-induced constriction of the rat aorta

Rats (SD strain, male, 10 to 14 weeks old) were bled to death under ether anesthesia and the thoracic aorta (ca 3 cm) was excised. The fat and connective tissue were removed and an annular specimen about 3 mm in width was prepared. The lumen was then abraded to remove vascular endothelial cells. This specimen was mounted on the isometric transducer of a Magnus tank filled with an organ bath and a static tensile load of 1 g was applied. The organ bath in the Magnus tank was maintained at 37°C under aeration with a mixed gas (95% O₂ + 5% CO₂), and with the organ bath changed at intervals of about 20 minutes, the specimen was allowed to equilibrate for about an hour. To this specimen was added calcium ionophore A23187 at a final concentration of 1 μM, and after the contractile response had steadied, the test compound was cumulatively administered. The relaxation response during the period was monitored and recorded and the 50% inhibitory concentration against A23187-induced vasoconstriction [IC₅₀ (μM)] was calculated. The IC₅₀, thus found, of the compound of Example 2 was 1.7. On the other hand, the IC₅₀ values of reference drugs, i.e. Compound A and fasudil

hydrochloride, were 10.6 and 7.4, respectively. The composition of the organ bath used in the experiment was: NaCl 115.9 mM (the same applies below); KCl 5.9; CaCl₂ 2.5; MgCl₂ 1.2; NaH₂PO₄ 1.2; NaHCO₃ 25.0; glucose 11.5; dissolved in distillation-deionized water. The pH of the bath saturated with the mixed gas was 7.4.

The compound of the invention has the property to relax calcium ionophore-induced vasoconstriction and the intensity of this action was significantly great as compared with the reference drugs, i.e. Compound A and fasudil hydrochloride.

Test Example 2

Rat middle cerebral arterial blood flow-increasing action

Using rats (SD, male, 11 to 12 weeks old) under urethane anesthesia, the head of each animal was immobilized and the buccinator muscle was peeled off to expose the cheek bone. Using a dental electric drill, an orifice about 5 mm in diameter was drilled in the cranium immediately over the middle cerebral artery (MCA) to provide for direct visual access to MCA. The probe (dia. 1.0 mm) of a laser Doppler blood flowmeter was brought close to the MCA to measure changes in MCA blood flow. The test compound was dissolved and diluted using saline and administered in a dose of 3 mg/kg from

the femoral vein through a cannula over 5 minutes. The action of each compound was expressed in terms of the percent increase in blood flow against the baseline value prior to administration. As a result, the compound of Example 2 increased the blood flow by 18.8%. In contrast, Compound A and fasudil hydrochloride, reference drugs, increased the flow only by 12.4% and 5.2%, respectively. In a further experiment, the test compound was administered over 30 minutes and the increase in blood flow was similarly measured. As a result, whereas the compound of Example 2 increased the blood flow significantly, by 5.2% at 1 mg/kg and 9.6% at 3 mg/kg, fasudil hydrochloride caused no increase at 3 mg/kg and a significant increase of 5.4% only at 10 mg/kg.

The compound of the invention thus has the property to increase the rat middle cerebral arterial blood flow and the intensity of this action was significantly great as compared with the reference drugs, i.e. Compound A and fasudil hydrochloride.

Test Example 3

Cerebral vasospasm-relieving action in the canine model of subarachnoid hemorrhage

In accordance with the method of Varsos et al. [J. Neurosurgery 58, 11-17 (1983)], the canine disease model was constructed by two successive injections of

autologous cisternal blood. On day 1 of experiment, with the animal under pentobarbital anesthesia, basilar artery angiography was taken prior to administration. Then, 4 ml of cerebrospinal fluid was removed from the cisterna magna and the same amount of autologous blood was injected at a rate of 2 ml/min. After blood injection, the head was tilted down through 30 degrees for 30 minutes so as to let the blood be uniformly distributed from the basilar artery to the Willis' cords. On day 3 of experiment, 4 ml of autologous blood was injected again into the cisterna magna. On day 7 of experiment, cerebral angiography was taken under pentobarbital anesthesia to check induction of delayed cerebral vasospasm and the efficacy evaluation was then carried out. The test compound was administered from the left femoral vein over 1 minute. Angiography was performed serially. The action of each compound was expressed in terms of the maximum diameter at the most constricted site of the basilar artery with the diameter at the same site on day 1 of experiment being taken as 100%.

As a result, against vasospasm, the compound of Example 2 exerted a significant relieving action at 0.3 mg/kg and higher doses and a complete relief of the vasospasm at 3 mg/kg. The blood pressure did not change at 0.3 mg/kg but was depressed at 1 mg/kg and higher

doses. On the other hand, fasudil hydrochloride showed a significant relieving action only immediately after drip injection of 10 mg/kg. The blood pressure fell at 3 mg/kg and higher doses.

Thus, assuming that the group treated with 3 mg/kg of the compound of Example 2 is Test Group A (the number of animals (n)=5), the group treated with 3 mg/kg of the reference drug fasudil hydrochloride is Test Group B (n=4), and the group treated with 10 mg/kg of fasudil hydrochloride is Test Group C (n=4), the diameter of the artery prior to medication on day 7 was 58% in Test Group A. The values in Test Groups B and C were 58% and 57%, respectively. The compound of Example 2, when administered intravenously, dilated the affected blood vessel to the caliber prior to blood injection, i.e. 107%. On the other hand, fasudil hydrochloride failed to show a definite relieving action at 3 mg/kg i.v. and relieved the vasospasm only to 82% even at 10 mg/kg i.v.

The compound of the invention, thus, relieved vasospasm significantly at a very low dose of 0.3 mg/kg not affecting the systemic blood pressure.

Test Example 4

Action on the rat cerebral infarction caused by transient middle cerebral arterial occlusion

Using male SD rats (210-240 g, aged 7 weeks, 12

animals per group) under halothane anesthesia, the common carotid artery was incised and a nylon obturator was inserted from the incision through the internal carotid artery to the origin of the middle cerebral artery. After this arrest of middle arterial blood flow, the anesthesia was stopped. After 2 hours, the nylon obturator was withdrawn for reperfusion. After 6 hours of reperfusion, the brain was enucleated and the infarcted region was identified by TTC (2,3,5-triphenyl-tetrazolium chloride) staining. The infarction volume was measured separately for the cerebral cortex and the superior brain stem region other than the cortex and Dunnett's test was used for statistical evaluation. The compound of Example 2, 0.3 and 1 mg/kg, or fasudil hydrochloride, 3 mg/kg, was administered by intravenous drip injection over 30 minutes starting immediately after the middle cerebral artery occlusion-reperfusion procedure.

As a result, the compound of Example 2 at 1 mg/kg was found to significantly inhibit the cerebral cortex infarction following transient middle cerebral arterial occlusion from 298.7 mm³ to 181.7 mm³ (P<0.05). On the other hand, fasudil hydrochloride at 3 mg/kg showed only a tendency toward inhibition of the brain infarction.

Test Example 5

Action on the rat cerebral infarction caused by permanent middle cerebral arterial occlusion

Using male SD rats (210-240 g, aged 7 weeks, 10 to 11 animals per group) under halothane anesthesia, a small orifice was drilled in the base region of the temporal bone and the left middle cerebral artery trunk was cauterized by electrocoagulation. At 48 hours after this middle cerebral artery occlusion, the brain was enucleated and the infarcted region was identified by TTC staining. The infarction volume was measured separately for the cerebral cortex and the striate body and Dunnett's test was used for statistical evaluation. The compound of Example 2, 0.03, 0.1, 0.3, or 1 mg/kg, or fasudil hydrochloride, 1 or 3 mg/kg, was administered by intravenous drip infusion over 30 minutes starting immediately after the middle cerebral arterial occlusion. As a result, in the cerebral cortex and striate body, the compound of Example 2 at 0.3 mg/kg suppressed the infarction volume significantly ($P < 0.01$) from 341.0 mm³ to 198.0 mm³ and from 141.8 mm³ to 103.8 mm³, respectively. However, fasudil hydrochloride suppressed the infarction volume significantly ($P < 0.01$) only at 3 mg/kg, from 341.0 mm³ to 224.7 mm³ in the cerebral cortex and from 141.8 mm³ to 103.7 mm³ in the striate body. Thus, the compound of Example 2 is more than 10 times as potent

as fasudil hydrochloride.

Test Example 6

Action on the rat cerebral infarction due to
photochemically induced thrombus

Using male SD rats (aged 7 weeks, 8 animals per group), an orifice was drilled in the skull immediately superior to the middle cerebral artery for insuring direct visual access to the middle cerebral artery. Rose Bengal was administered intravenously and a light source for construction of a thrombus model was brought close to the middle cerebral artery for exposure to green light. Immediately after exposure to green light, the test drug was administered intravenously. After 24 hours the brain was enucleated and a continuous series of coronary sections was prepared and stained with TTC solution. As a result, the compound of Example 2 was found to inhibit cerebral infarction in both the cerebral cortex and the striate body, with the effect being more remarkable in the cerebral cortex. Thus, at the doses of 0.3 and 1 mg/kg, the compound suppressed the infarction volume in the cerebral cortex significantly from 86.3 mm³ to 64.5 mm³ and 55.6 mm³ ($P < 0.05$, $P < 0.01$), respectively. At 1 mg/kg, the compound suppressed the infarction volume in the striate body significantly from 53.2 mm³ to 37.3 mm³ ($P < 0.05$). In contrast, fasudil hydrochloride

suppressed the infarction volume in the striate body significantly from 59.9 mm³ to 43.5 mm³ and 44.5 mm³ only at the doses of 1 and 3 mg/kg, respectively ($P < 0.01$).

Thus, the compound of Example 2 was found to be considerably more effective than fasudil hydrochloride in relieving cerebral infarction of the thrombus type.

Test Example 7

Action of the compound of Example 2 on coronary arterial blood flow in anesthetized open-chest dogs

Protocol

Beagle dogs weighing 12.3-13.4 kg (in groups of 4 animals) were anesthetized with pentobarbital sodium (30 mg/kg i.v.) and immobilized in supine position. Thereafter up till the end of the experiment, pentobarbital sodium (3-5 mg/kg/hr) was infused continuously from a catheter indwelled in the right cephalic vein. After tracheal intubation and introduction of supportive respiration with an artificial respirator (tidal volume 15 ml/kg, respiration frequency 20 beats/min), a left thoracotomy was performed in the interspace between the 4th and 5th ribs, the pericardium was incised, and the heart was suspended in the manner of a hammock. The probe of an electromagnetic blood flowmeter was mounted on the anterior descending branch of the left coronary artery

and using the electromagnetic flowmeter and a bioelectric preamplifier, the coronary arterial blood flow was measured and recorded on a Recticorder. The compound of Example 2 and fasudil hydrochloride were respectively dissolved extemporaneously in saline in a dose volume of 30 ml per animal and infused over 30 minutes via the catheter indwelled in the left cephalic vein. The % change from the baseline value before the start of infusion was calculated. Measurement of blood flow was performed over 60 minutes inclusive of the dosing time. As a result, in the group treated with saline, little change was found in coronary arterial blood flow. The time course of coronary arterial blood flow after the start of intravenous administration of the drug substance was recorded. After 5 to 15 minutes after the start of administration of the compound of Example 2, the blood flow was increased significantly by 29.7%, 69.3%, and 86.7% at 0.1, 0.3, and 1 mg/kg, respectively. On the other hand, with the reference compound fasudil hydrochloride, only a tendency toward increase was found at 3 mg/kg and a significant increase of 53.1% only at 10 mg/kg.

It is apparent from Test Examples 1 to 7 that the compound of the invention showed remarkably superior cerebral vasospasm-relieving activity and

blood-flow-increasing activity even compared with the known compound which is structurally analogous to the compound of the invention.

Test Example 8

Acute toxicity

The test was performed in rats in accordance with the method of Wyle. Using 6-year-old male slc SD rats (5 animals/group), the test drug was administered from the tail vein over 60 seconds and the mortality rate within 24 hours was investigated. The test drug was used as dissolved and diluted with saline. As a result, the acute toxicity of the compound of Example 2 was found to be low.

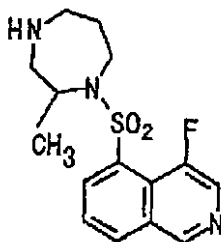
INDUSTRIAL APPLICABILITY

The compound of the invention thus showed a potent cerebral vasospasm-relieving action at a considerably lower dose not affecting the blood pressure as compared with the structurally analogous reference Compound A and the reference compound fasudil hydrochloride. Furthermore, the compound of the invention restored the caliber of the constricted blood vessel completely to the caliber before blood injection. The findings suggested that the compound of the invention is useful for the prevention and treatment of brain tissue damages due to cerebrovascular disease, particularly cerebral

vasospasm secondary to cerebral hemorrhage. In addition to the above-mentioned cerebral vasospasm-relieving action, the compound of the invention has organ-selective cerebral and cardiac blood vessel-dilating activity, blood flow-increasing activity and ischemic neuron-protecting activity at doses considerably lower the corresponding doses of the reference compound fasudil hydrochloride and, therefore, is of value as a prophylactic or therapeutic drug for diseases accompanied by ischemic lesions. Since the compound of the invention dilates the cerebral blood vessels and displays cerebral blood flow-increasing and ischemic neuron-protecting activities, it is of value as a prophylactic or therapeutic drug for sequelae (e.g. dyskinesia) of cerebral hemorrhage, cerebral infarction, transient cerebral ischemic attack, and head trauma. In addition, because it dilates the coronary blood vessels to show a potent coronary blood flow-increasing action, the compound is useful for the prevention and treatment of myocardial infarction and angina pectoris.

CLAIMS

1. A compound of the following general formula [I] or a medically acceptable salt thereof.



[I]

2. The compound or pharmaceutically acceptable salt of Claim 1 which has an absolute configuration of S.
3. A pharmaceutical composition comprising the compound or medically acceptable salt of Claim 1 as an active ingredient.
4. The pharmaceutical composition of Claim 3 which is a prophylactic or therapeutic drug for cerebrovascular disease.
5. The pharmaceutical composition of Claim 3 which is a prophylactic or therapeutic drug for diseases accompanied by ischemic lesions.
6. The pharmaceutical composition of Claim 3 which is a cerebral vasospasm inhibitor.
7. The pharmaceutical composition of Claim 3 which is a protectant for ischemic neuronal death.
8. The pharmaceutical composition of Claim 3

wherein the disease type of said cerebrovascular disease is cerebral infarction.

9. The pharmaceutical composition of Claim 3 wherein the disease type of said cerebrovascular disease is transient cerebral ischemic attack.

10. The pharmaceutical composition of Claim 3 wherein the disease types of said cerebrovascular disease are cerebral hemorrhage and head trauma.

11. The pharmaceutical composition of Claim 3 which is a prophylactic or therapeutic drug for sequelae of cerebral hemorrhage and head trauma.

12. The pharmaceutical composition of Claim 3 which is a prophylactic or therapeutic drug for ischemic heart disease.

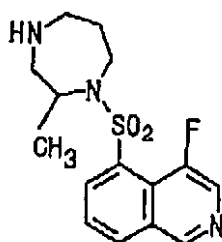
13. The pharmaceutical composition of Claim 3 which is a myocardial infarction inhibitor.

14. The pharmaceutical composition of Claim 3 which is a therapeutic drug for angina pectoris.

ABSTRACT

The invention relates to an isoquinoline derivative of the following general formula [I] or a medicinally acceptable salt thereof and to a pharmaceutical composition containing any of them as an active ingredient.

The compound [I] is useful as a prophylactic or therapeutic drug for cerebrovascular disease, particularly as a compound of great value as a cerebral vasospasminhibitor. It is also useful as a prophylactic or therapeutic drug for diseases accompanied by ischemic lesions.



[I]