R₂, when considered together with R₃, represents an additional bond between the carbon atoms to which R₂ and R₃ are attached or R₂ in case Z represents the group (b) a hydrogen atom.
We, GRUNENTHAL GMBH, of 5190 Stolberg/Rhld, Federal Republic of Germany hereby apply for the grant of a standard patent for an invention entitled:

"USE OF DIPEPTIDE DERIVATIVES IN THE MANUFACTURE OF MEDICAMENTS FOR THE TREATMENT OF POSTTRAUMATIC NERVOUS INJURIES"

which is described in the accompanying complete specification.

DETAILS OF BASIC APPLICATION

Number of Basic Application:-
P 35 02 041.5

Name of Convention Country in which Basic Application was filed:-
Federal Republic of Germany

Date of Basic application:-
23 January, 1985

Our address for service is:-
C/- Spruson & Ferguson
Patent Attorneys
Level 33 St Martins Tower
31 Market Street
Sydney New South Wales Australia

DATED this SEVENTH day of JANUARY 1986

GRUNENTHAL GMBH

By:


TO: THE COMMISSIONER OF PATENTS
AUSTRALIA
In support of the Convention Application made for a patent for an invention entitled:

"Use of dipeptide derivatives in the manufacture of medicaments for the treatment of posttraumatic nervous injuries"

We, Dr. Franz Wirtz and Dr. Siegfried Herrling

of Atzenach 37, D-5190 Stolberg, Fed. Rep. of Germany

and of Dohnenweg 33, D-5190 Stolberg, Fed. Rep. of Germany, respectively,

do solemnly and sincerely declare as follows:

1. I/we, we are the applicant(s) for the patent

(or, in the case of an application by a body corporate)

1. I/we are authorised by Grüenthal GmbH (D-5190 Stolberg)

the applicant(s) for the patent to make this declaration on its/their behalf.

2. The basic application(s) as defined by Section 141 of the Act was/were made

in Federal Republic of Germany

on January 23, 1985

by Grüenthal GmbH

3. I/we, we are the actual inventor(s) of the invention referred to in the basic application(s)

(or where a person other than the inventor is the applicant)

3. Prof. Dr. Hubert Giertz

Priv. Doz. Dr. Hans Barth and

Prof. Dr. Leopold Flohe


and of Taubengasse 12, D-5100 Aachen, Fed. Rep. of Germany

and of Lenzbachstrasse 24, D-5106 Roetgen, Fed. Rep. of Germany

(respectively)

are the actual inventor(s) of the invention and the facts upon which the applicant(s) is/are entitled to make the application are as follows:

The inventors are employees of the applicant and the right to make the application was acquired by the applicant under the German law on inventions of employees.

4. The basic application(s) referred to in paragraph 2 of this Declaration was/were the first application(s) made in a Convention country in respect of the invention(s) the subject of the application.

Declared at Stolberg this 13 day of November 1985

[Signatures of Declarant(s)]
Claim

1) Use of dipeptide derivatives of the form

\[
\text{Z} \begin{array}{c}
\text{N} \\
\text{C}
\end{array} \text{C} \begin{array}{c}
\text{N} \\
\text{H}
\end{array} \text{O} \begin{array}{c}
\text{N} \\
\text{H}
\end{array} \text{CH} \begin{array}{c}
\text{H} \\
\text{H}
\end{array} \text{N} \begin{array}{c}
\text{H} \\
\text{H}
\end{array} \text{CO-NH-CH-CO-N} \text{CO-NHR}_1
\]

wherein

\( R_1 \) represents a hydrogen atom, an alkyl radical containing one to six carbon atoms, a cyclohexyl group or a benzyl group,

\( Z \) is one of the following groups (attached to the CO-group in the ring by the valence marked with an asterisk)

\[
(a) \equiv \text{NH-CO-} \quad \text{or} \quad (b) \equiv \text{S-CH}_2
\]

\[
\text{R}_3 \quad \text{R}_4 \quad \text{R}_5 \quad \text{R}_6
\]

.../2
$\text{R}_2$, when considered together with $\text{R}_2$, represents an additional bond between the carbon atoms to which $\text{R}_2$ and $\text{R}_3$ are attached or $\text{R}_2$ is, in case $Z$ represents the group (b), a hydrogen atom.

$\text{R}_4$ and $\text{R}_5$ are equal or different and each represents hydrogen or an alkyl radical containing one to three carbon atoms, $\text{R}_5$ also may represent a phenyl group, and $\text{R}_6$ represents a hydrogen atom or a methyl group and of pharmaceutically acceptable acid addition salts of these compounds for the manufacture of medicaments for the treatment of posttraumatic nervous injuries.
an additional bond are attached or \( R_2 \)
drogen atom,
ents hydrogen or
ee carbon atoms, and

ents of these
ent of posttraumatic
ent of posttraumatic

Complete Specification

FORM 10

COMMONWEALTH OF AUSTRALIA

PATENTS ACT 1952

COMPLETE SPECIFICATION

(ORIGINAL)

FOR OFFICE USE:

Spruson & Ferguson

Class Int. Class

Complete Specification Lodged:

Accepted:

Published:

Priority:

Related Art:

Name of Applicant: GRUNENTHAL GMBH

Address of Applicant: 5190 Stolberg/Rhld, Federal Republic of Germany

Actual Inventor(s): HUBERT GIERTZ, HANS BARTH and LEOPOLD FLOHE

Address for Service: Spruson & Ferguson, Patent Attorneys, Level 33 St Martins Tower, 31 Market Street, Sydney, New South Wales, 2000, Australia

Complete Specification for the invention entitled:

"USE OF DIPEPTIDE DERIVATIVES IN THE MANUFACTURE OF MEDICAMENTS FOR THE TREATMENT OF POSTTRAUMATIC NERVOUS INJURIES"

The following statement is a full description of this invention, including the best method of performing it known to us

SBR/TGR/193U
ABSTRACT

Dipeptide derivatives of the formula

\[
\begin{align*}
\text{O} & \text{C} \text{N} \text{H} \\
\text{C} & \text{O-NH-CH-CO-N} \\
& \text{CO-NHR}_1
\end{align*}
\]

wherein \( R_1 \) is hydrogen, a lower alkyl group, cyclohexyl or benzyl, \( Z \) is one of the groups

\[
\begin{align*}
\text{N} & \text{H} \\
\text{c} & \text{H}_2 \\
\text{c} & \text{c} \text{-CH} \text{-CO-NHR}_1
\end{align*}
\]

(a) \( \text{NH-CO-C} \) or (b) \( \text{C-S-CH}_2 \)

\( \text{R}_2 \), when considered together with \( R_3 \), represents an additional bond between the carbon atoms bearing \( R_2 \) and \( R_3 \) or \( R_2 \) is, if \( Z \) represents the group (b), hydrogen, \( R_4 \) and \( R_5 \) are hydrogen or lower alkyl, \( R_5 \) may also be phenyl, and \( R_6 \) is hydrogen or methyl and pharmaceutically acceptable acid addition salts of these compounds are used in the manufacture of medicaments for prevention or treatment of the sequels of traumatic nervous injuries especially of traumatic injuries of the spinal cord and/or the brain. These medicaments are suitable for the application of the active ingredient(s) to the patient preferably in the initial treatment by infusion or injection. For the subsequent treatment the medicament also may administered orally, perorally, rectally or percutaneously.
Use of dipeptide derivatives in the manufacture of medicaments for the treatment of posttraumatic nervous injuries.

Hurts of the spinal cord and/or of the brain relatively often occur in traffic or sports accidents. Depending on the degree or the severity, respectively, of these traumata more or less pronounced symptoms develop (such as paraplegia or paralysis due to spinal cord injuries or central paralysis, impairment of the memory and other neurological deficiencies due to traumata of the brain, respectively) which often are irreversible and in any case are difficult to cure. Recently it was described that certain compounds as for instance thyrotropin-releasing-hormone (TRH = L-pyroglutamyl-L-histidyl-L-prolinamide) in animal experiments significantly improve recovery of the motoric function after experimental spinal cord injury when treatment occurs during the first 24 hours following the trauma. Other authors observed a dose-dependent positive effect of TRH on EEG alterations resulting in animal experiments from traumata of the brain (caused by brain stem compression in cats). Furthermore it has been described that impaired consciousness in humans due to functional or organic damage of the brain such as cranial trauma, cerebrovascular disorder, cerebral surgery and brain tumor may be improved by administration of TRH successively for at least 10 days.

Unfortunately in the living organism TRH is quickly metabolized and inactivated by enzymatically splitting off the pyroglutamyl group and/or deamidation. Accordingly TRH or its salts are generally administered by continuous intravenous infusions in high doses and in exceptional cases only intramuscular or peroral administration may be taken into consideration.
It has been tried, already, to synthesize by chemical modification of the TRH-molecule compounds which have the desired biological activities of TRH but which are stable against the metabolizing and inactivating enzymes thus being avoid of the disadvantages of TRH resulting from its instability. These TRH-modifications show, however, different profiles of biologic activity and especially these activities are only in part equivalent to those of TRH. These results were ascertained, however, only with respect to the endocrinological effects and to the stimulating effects on the central nervous system. Accordingly it is completely unknown and unpredictable whether such TRH-modifications have any effects - and, if so, which effects they may have - for instance on posttraumatic nervous injuries, especially on paraplegia due to spinal cord injuries and/or on neurological deficiencies after traumata of the brain.

It now surprisingly has been found that by administration of medicaments containing dipeptide derivatives of the formula

\[
\begin{align*}
\text{I} & \quad \text{wherein} \\
R_1 & \text{represents a hydrogen atom, an alkyl radical containing one to six carbon atoms, a cyclohexyl group or a benzyl group,} \\
Z & \text{is one of the following groups (attached to the CO-group in the ring by the valence marked with an asterisk)} \\
R_3 & \text{or } R_5 \\
R_4 & \text{or } R_6
\end{align*}
\]
R₂, when considered together with R₃, represents an additional bond between the carbon atoms to which R₂ and R₃ are attached or R₂ is, in case Z represents the group (b), a hydrogen atom,

R₄ and R₅ are equal or different and each represents hydrogen or an alkyl radical containing one to three carbon atoms and wherein R₅ also may represent a phenyl group and

R₆ represents a hydrogen atom or a methyl group

and pharmaceutically acceptable acid addition salts of these compounds

the symptoms due to traumata of the spinal cord and/or of the brain can be prevented or treated successfully.

Preferred compounds of formula I to be used in the manufacture of medicines for the treatment of posttraumatic nervous injuries, especially in the prevention or treatment, respectively, of paraplegia or paralysis due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain are those of the formulae

![Chemical Structures](image-url)
wherein \( R_1 \) and \( R_5 \) have the same meaning as above.

Preferably in formulas I, Ia and Ib, respectively, \( R_1 \) represents a hydrogen atom.

Especially preferred members of the compounds of formula I are orotyl-L-histidyl-L-prolinamide and 5-oxo-6-methyl-thiomorpholine-3(L)carbonyl-L-histidyl-L-prolinamide.

The compounds of formula I and their preparation have been described e. g. by Schwértner et al. in "Structure and Activity of Natural Peptides" (Editors W. Voelter and G. Weitzel) Walter de Gruyter-Verlag, Berlin - New York 1981, pages 397 - 415, in German patents 2.449.167 and 2.615.455, and elsewhere.

In the cited publications it has been shown that the compounds of formula I on parenteral or oral administration provide long lasting central stimulating effects and that the toxicity of the compounds is very low. Due to these pharmacological properties the compounds of formula I according to the publications mentioned above and further references can be used as psycho-stimulating agents or as anti-depressive agents, respectively.

The known fields of indications for the compounds of formula I accordingly relate to chronic mental illness and not to acutely occurring neurological deficiencies. Therefore it was unforeseeable that the compounds of formula I or medicaments containing them, respectively, can be used for the treatment of posttraumatic nervous injuries, especially to prevent or to treat, respectively, paraplegia due to spinal cord injuries and/or neurological deficiencies after traumata of the brain.

Accordingly the present invention relates to the use of dipeptide derivatives of formula I and of their pharmaceutically acceptable acid addition salts for the manufacture of medicaments for the treatment of posttraumatic
nervous injuries of the types mentioned above, especially for prophylaxis or therapy of posttraumatic nervous injuries caused by accidents, such as paraplegia and/or neurological deficiencies.

If possible the first dose of a medicament containing a compound of formula I to patients suffering from central nervous traumata should be administered already at the place of accident, preferably in form of a preparation suitable for injection containing for instance 5 to 100 mg of the active ingredient of formula I or an acid addition salt thereof. The subsequent treatment is made in the hospital by administering the medicament in form of intravenous or intrathecal infusions or in form of intravenous, intramuscular, subcutaneous or intraperitoneal injections, respectively.

Such preparations suitable for infusions or injections are known per se and consist, for instance, in a bottle closed with a rubber stopper and containing the desired amount of the active compounds in sterile, dry (lyophilized) form. By adding a suitable solvent such as, for instance, sterile isotonic aqueous solutions of sodium chloride, glucose, inositol, mannitol or the like, in which the compounds of formula I are readily soluble, and adjusting the resulting solution (which may be used as it is for injections) to the appropriate volume by adding it to the desired amount of the same solvent in an infusion flask a solution to be used for infusions is obtained.

Very useful application forms are spray forms for intranasal or oral applications of the compounds of formula I or for the administration of these substances to the bronchia. Such sprays can be prepared as known from the state of the art.

The compounds of formula I and their salts are relatively stable products. Thus their incorporation into pharmaceutical compositions causes no problems for the skilled pharmacist. The following explanations and examples are given for illustration only:
Especially to maintain the plasma level resulting from the introductory treatment with infusions or injections, respectively, oral or peroral application forms of the compounds of formula I are useful. Such application forms like tablets, dragees, capsules, granules, drops and sirups are known per se. In their production generally used inorganic or organic adjuvants such as diluents, carriers, binders, lubricants, colours, flavorings and others are added to the compounds of formula I.

For instance tablets or dragees, each containing 20 mg of a compound of formula I, may be prepared by mixing 20 g of the respective active ingredient of formula I together with 35 g of corn starch, 10 g of colloidal silica, 5 g of magnesium stearate, and, if desired, colours and/or flavorings. The blend is granulated, dried and compressed into 1000 tablets which may become film-coated or sugar-coated.

Capsules, each containing 20 mg of the compound of formula I, may be prepared for instance by mixing 20 g of the active ingredient with 376 g of lactose, granulating the mixture with an aqueous solution of 4 g gelatine, drying and finally filling into 1000 hard-shell gelatine capsules.

Drops for intranasal application, which may be used also in form of a spray, can be obtained in a manner known per se by dissolving the compound of formula I in an isotonic aqueous solution of sodium chloride, mannitol, sorbitol, inositol or the like and adding an adhesive like polyvinyl pyrrolidone or polyvinyl alcohol and/or a preservative like 4-hydroxybenzotric acid methyl ester or benzyl alcohol.

Suppositories containing an active ingredient of formula I may be prepared by melting 95 g of a commercially available suppositories base at about 40 - 45°C, adding 3 g of salicylic or mandelic acid, followed by adding, while stirring, 2 g of the active ingredient and pouring the mixture into moulds.
Very convenient are in several cases also compositions for percutaneous application of the compounds of formula I, such as plasters or the like containing a solution of the active ingredient, optionally also containing a known membrane penetration enhancer such as a N-alkyl lactam etc.

The pharmaceutical compositions mentioned above for peroral, rectal, percutaneous or intramuscular administration of the compounds of formula I preferably may be such from which the active ingredient has a delayed release. Thus for a longer period of time, for instance 24 hours, a steady supply of the patient with the active ingredient can be reached.

The amounts of the compounds of formula I to be administered to prevent or to treat the symptoms after spinal cord or brain traumata depend - besides their application form and place - on the degree of the injuries in the individual patient and are to be determined individually by the physician. In general it is advisable to administer about 1 to 300 mg per die by infusion or by injection, respectively.

The surprising activity of the compounds of formula I and of medicaments containing these ingredients in the treatment of spinal cord injuries was demonstrated in animal experiments as follows:

Cats anesthetized with sodium pentobarbital were subjected under aseptic conditions to a laminectomy to expose the C-7 spinal segment. With the dura intact, the spinal cord was traumatized by dropping a 20 g weight a distance of 30 cm through a guide tube onto a 10 square-millimeter impact plate. Drugs were given intravenously as a bolus injection at 1 and 3 hours after injury. A group of 9 animals was treated with 0.2 mg/kg of orotyl-L-histidyl-L-prolinamide (a compound of formula I), dissolved in 0.5 ml of physiologic saline. A second group of 7 animals was given 1 mg/kg L-pyro-2-aminoadipyl-L-histidyl-thiazolidine-4-carboxamide (also dissolved in 0.5 ml of physiologic saline), which compound hereinafter is referred to as "compound A" and which often in the literature is described as a TRH-modification product and is compared with the orotyl-L-
histidyl-L-prolinamide mentioned before (c.f. Metcalf in "Thyrotropin Releasing Hormone", editors E.C. Griffiths and G. W. Bennet, Raven Press, New York 1983, pages 315 to 326). A control group of 11 animals was treated with 0.5 ml of physiologic saline per animal.

Neurologic function was evaluated weekly for six weeks and assessed according to a predetermined scale. Thereby it could be found that after 6 weeks the neurologic and the motoric function in the animals treated with orotyl-L-histidyl-L-prolinamide were significantly improved whereas compound A or saline, respectively, had no significant effect (although the dose of compound A administered was 5 times higher than that of orotyl-L-histidyl-L-prolinamide). These results also prove, that the effect of a TRH-modification product on the central nervous system (compound A has such effect in a pronounced degree) has no relationship to an effect on the activity of said product to prevent or cure posttraumatic nervous injuries (as demonstrated in the experiment described above in the model of paraplegia due to spinal cord injuries) i.e. that the observed significant activity of orotyl-L-histidyl-L-prolinamide in this experiment could neither be expected nor predicted on the basis of the known pharmacological properties of this compound of formula I.

Results similar to those obtained by the treatment using orotyl-L-histidyl-L-prolinamide were also observed on administration of other compounds of formula I in form of respective pharmaceutical compositions. The effective doses in these treatments were, for instance, 0.1 mg/kg, 0.2 mg/kg, 0.5 mg/kg and 1 mg/kg. Due to the known low toxicity of the compounds of formula I it is also possible to use still higher doses and/or to administer the desired amount of the compound of formula I several times a day (e.g. 3 to 5 times daily) even for several consecutive days up to several weeks.

Effects comparable to the results obtained in the animal experiments described above were also observed by administering medicaments containing a compound of formula I to victims of spinal-cord injuries due to accidents in which the degree of the injury most probably otherwise would
it was from ethical and other reasons naturally impossible to perform control experiments for instance by administering the (in animal experiments ineffective) compound A or saline only.
The claims defining the invention are as follows:

1) Use of dipeptide derivatives of the formula

\[
\text{R}_1 \text{represents a hydrogen atom, an alkyl radical containing one to six carbon atoms, a cyclohexyl group or a benzyl group,}
\]

\[Z \text{is one of the following groups (attached to the CO-group in the ring by the valence marked with an asterisk)}
\]

\[
\begin{align*}
\text{(a) } & \text{NHCO-} & \text{(b) } & \text{SCH}_2
\end{align*}
\]

\[
\text{R}_2, \text{when considered together with } \text{R}_3, \text{represents an additional bond between the carbon atoms to which } \text{R}_2 \text{and } \text{R}_3 \text{are attached or } \text{R}_2 \text{is, in case } Z \text{represents the group (b), a hydrogen atom,}
\]

\[
\text{R}_4 \text{and } \text{R}_5 \text{are equal or different and each represents hydrogen or an alkyl radical containing one to three carbon atoms,}
\]

\[
\text{R}_5 \text{also may represent a phenyl group, and}
\]

\[
\text{R}_6 \text{represents a hydrogen atom or a methyl group}
\]

and of pharmaceutically acceptable acid addition salts of these compounds

-10-
for the manufacture of medicaments for the treatment of posttraumatic nervous injuries.

2) Use of dipeptide derivatives of formula I of claim 1 and of pharmaceutically acceptable acid addition salts of these compounds for the manufacture of medicaments for prevention or treatment of paraplegia or paralysis due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain.

3) Use of dipeptide derivatives of formula I of claim 1 and of pharmaceutically acceptable acid addition salts of these compounds for the manufacture of medicaments for prevention or treatment of posttraumatic nervous injuries resulting from spinal cord injuries and/or traumata of the brain caused by accidents.

4) Use of dipeptide derivatives of formula I of claim 1 and of pharmaceutically acceptable acid addition salts of these compounds for the manufacture of medicaments for prevention or treatment of paraplegia or paralysis due to spinal cord injuries caused by accidents.

5) Use of dipeptide derivatives of the formula

\[
\begin{align*}
  &
  \text{wherein } R_2 \text{ and } Z \text{ have the same meaning as defined in formula I} \\
  &\text{and of pharmaceutically acceptable acid addition salts of these compounds}
\end{align*}
\]
The following statement is a full description of this invention, including the best method of performing it known to us.

5) Use of dipeptide derivatives of the formula

\[ \text{Formula I} \]

wherein \( R_1 \) is defined in formula I

and of pharmaceutically acceptable acid addition salts of these compounds

for the manufacture of medicaments for the treatment of posttraumatic nervous injuries, especially for prevention or treatment of paraplegia due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain.

7) Use of dipeptide derivatives of the formula

\[ \text{Formula II} \]

wherein \( R_1 \) and \( R_5 \) have the same meaning as defined in formula I.
and of pharmaceutically acceptable acid addition salts of these compounds for the manufacture of medicaments for the treatment of posttraumatic nervous injuries, especially for the prevention or treatment of paraplegia due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain.

8) Use of orotyl-L-histidyl-L-prolinamidc or of a pharmaceutically acceptable acid addition salt of this compound for the manufacture of medicaments for the treatment of posttraumatic nervous injuries, especially for the prevention or treatment of paraplegia due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain.

9) Use of 5-oxo-6-methyl-thiomorpholine-3-(L)-carbonyl-L-histidyl-L-prolinamide or of a pharmaceutically acceptable acid addition salt of this compound for the manufacture of medicaments for the treatment of posttraumatic nervous injuries, especially for the prevention or treatment of paraplegia due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain.

10) Use of dipeptide derivatives of the formulas given in claims 1 and 5 to 7 and of pharmaceutically acceptable acid addition salts of these compounds according to claims 1 to 9 for the manufacture of medicaments suitable for administration of the active ingredient by infusion or injection for the treatment of posttraumatic nervous injuries, especially for the prevention or treatment of paraplegia due to spinal cord injuries and/or of neurological deficiencies after traumata of the brain caused by accidents.

11) Use of dipeptide derivatives of the formulas given in claims 1 and 5 to 7 and of pharmaceutically acceptable acid addition salts of these compounds according to claims 1 to 9 for the manufacture of medica-


ments suitable for percutaneous administration of the active ingre-
dient for the treatment of posttraumatic nervous injuries, especially
for the prevention or treatment of paraplegia due to spinal cord
injuries and/or of neurological deficiencies after traumata of the
brain caused by accidents.

12) Use of dipeptide derivatives of the formulas given in claims 1 and
5 to 7 and of pharmaceutically acceptable acid addition salts of
these compounds for the manufacture of medicaments suitable for oral,
peroral or rectal administration for the treatment of posttraumatic
nervous injuries, especially for the prevention or treatment of para-
plegia due to spinal cord injuries and/or of neurological deficiencies
after traumata of the brain caused by accidents.

DATED this SEVENTH day of JANUARY, 1986

(RUNENTHAL GMBH

Patent Attorneys for the Applicant
SPRUSON & FERGUSON