

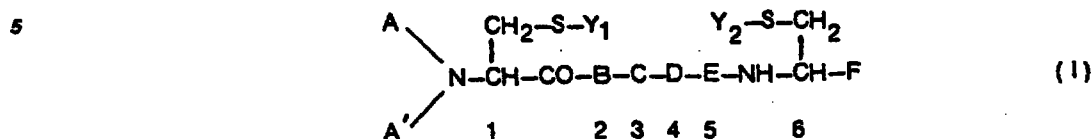
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Polypeptides, processes for their production, pharmaceutical compositions comprising said polypeptides and their use

The present invention provides novel polypeptides, processes for their production, pharmaceutical compositions comprising said polypeptides and their use as pharmaceutically active agents.

More particularly the present invention provides a polypeptide of formula I



wherein

A is C₁₋₁₂ alkyl, C₇₋₁₀ phenylalkyl or a group of formula RCO—, whereby

i) R is hydrogen, C₁₋₁₁ alkyl, phenyl or C₇₋₁₀ phenylalkyl, or

ii) RCO— is

15 a) an L- or D-phenylalanine residue optionally ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl and/or C₁₋₃ alkoxy

b) the residue of a natural α-amino acid other than defined under a) above or of a corresponding D-amino acid, or

20 c) a dipeptide residue in which the individual amino acid residues are the same or different and are selected from those defined under a) and/or b) above, the α-amino group of amino acid residues a) and b) and the N-terminal amino group of dipeptide residues c) being optionally mono- or di-C₁₋₁₂ alkylated,

A' is hydrogen or, when A is C₁₋₁₂ alkyl or C₇₋₁₀ phenylalkyl, also C₁₋₁₂ alkyl or C₇₋₁₀ phenylalkyl,

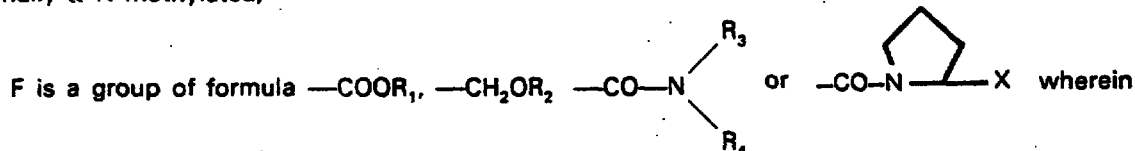
B is —Phe— optionally ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl and/or C₁₋₃ alkoxy,

25 C is —(L)— or —(D)—Trp— optionally α-N-methylated and optionally benzene-ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl and/or C₁₋₃ alkoxy,

D is —Lys— optionally α-N-methylated and optionally ε-N-C₁₋₃ alkylated,

E is the residue of a natural α-amino acid or of a corresponding D-amino acid, said residue being optionally α-N-methylated,

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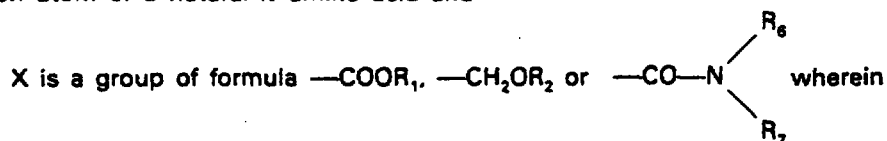
R₁ is hydrogen or C₁₋₃ alkyl,

R₂ is hydrogen or the residue of a physiologically acceptable, physiologically hydrolysable ester,

R₃ is hydrogen, C₁₋₃ alkyl, phenyl or C₇₋₁₀ phenylalkyl,

R₄ is hydrogen, C₁₋₃ alkyl or, when R₃ is hydrogen or methyl, also a group of formula —CH(R₅)—X,

40 R₅ is hydrogen, —(CH₂)₂—OH or —(CH₂)₃—OH, or represents the substituent attaching to the α-carbon atom of a natural α-amino acid and



R₁ and R₂ have the meanings given above,

R₆ is hydrogen or C₁₋₃ alkyl and

50 R₇ is hydrogen, C₁₋₃ alkyl, phenyl or C₇₋₁₀ phenylalkyl, the group —CH(R₅)—X having the D- or L-configuration, and

Y₁ and Y₂ are each hydrogen or together represent a direct bond, whereby the residues in the 1- and 6-position each independently have the L- or D-configuration, with the proviso that D- and/or L-cysteine residues are present at the 1- and 6-positions only, as well as the salt forms and complexes thereof. In the polypeptides of formula I, the following significances or combinations thereof are preferred:

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1. A is C₇₋₁₀ phenylalkyl, especially phenethyl, or a group of formula RCO. Preferably A is a group of formula RCO.

60 1.1. Preferably R is C₁₋₁₁ alkyl or C₇₋₁₀ phenylalkyl, especially C₇₋₁₀ phenylalkyl, more especially phenethyl, or

RCO has the meanings a), b) or c).

1.2. When RCO has the meanings a), b) or c), the α -amino group of amino acid residues a) and b) and the N-terminal amino group of dipeptide residues c) is preferably non-alkylated or mono-C₁₋₁₂ alkylated, especially -C₁₋₃ alkylated, more especially -methylated. Most preferably the N-terminal is non-alkylated.

1.3. When RCO has the meaning a) this is preferably a) an L- or D-phenylalanine or -tyrosine residue optionally mono-N-C₁₋₁₂ alkylated. More preferably a') is an L- or D-phenylalanine residue or an L- or D-N-(C₁₋₃-alkyl)-phenylalanine residue. Most preferably a') is a D-phenylalanine or D-N-(C₁₋₃ alkyl) phenylalanine residue, especially a D-phenylalanine or D-(N-methyl)-phenylalanine residue.

1.4. When RCO has the meaning b) or c) the defined residue is preferably lipophilic. Preferred residues b) are thus b') α -amino acid residues having a hydrocarbon side chain, e.g. leucine and nor-leucine residues, said residues having the L- or D-configuration, and preferred residues c) are dipeptide residues in which the individual amino acid residues are the same or different and are selected from those defined under a') and b') above.

1.5. Most preferably RCO has the meaning a), especially the meanings a').

2. B is -Phe-

3. C is -(D)Trp-

4. D is -Lys-, -MeLys- or -Lys(ϵ -Me)-, especially -Lys-.

5. E is the residue of a natural α -amino acid, especially -Thr-.

6. F is a group of formula $\text{—CO—N} \begin{matrix} \text{R}_3 \\ \text{R}_4 \end{matrix}$ especially a group of formula $\text{—CO—N} \begin{matrix} \text{R}_3 \\ \text{CH(R}_5\text{)—X} \end{matrix}$

(in which case R₃ = H or CH₃). In the latter case the moiety —CH(R₅)—X preferably has the L-configuration.

6.1. R₃ is preferably hydrogen.

6.2. As the substituent attaching to the α -carbon atom of a natural amino acid (i.e. of formula H₂N—CH(R₅)—COOH), R₅ is preferably —CH₂OH, —CH(CH₃)—OH, isobutyl or benzyl, or R₅ is —(CH₂)—OH or —(CH₂)₃—OH. It is especially —CH₂OH or —CH(CH₃)OH.

6.3. X is preferably a group of formula $\text{—CO—N} \begin{matrix} \text{R}_6 \\ \text{R}_7 \end{matrix}$

or —CH₂—OR₂ especially of formula —CH₂—OR₂ and R₂ is preferably hydrogen or has the meaning given under 7 below. Most preferably it is hydrogen.

7. As the residue of a physiologically acceptable, physiologically hydrolysable ester R₂ is preferably HCO, C₂₋₁₂ alkylcarbonyl, C₆₋₁₂ phenylalkylcarbonyl or benzoyl.

8. Preferably the residues in the 1- and 6-positions have the L-configuration.

9. Preferably Y₁ and Y₂ together represent a direct bond A group of compounds in accordance with the invention are polypeptides of formula I as defined above wherein

A is hydrogen, C₁₋₃ alkyl, C₇₋₁₀ phenylalkyl or a group of formula RCO—,

A' is hydrogen,

R is hydrogen, C₁₋₃ alkyl, phenyl or C₇₋₁₀ phenylalkyl or

RCO is a) an L- or D-phenylalanine residue optionally mono-ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl or C₁₋₃ alkoxy, or

b) the residue of a natural α -amino acid other than defined under a) above or of a corresponding D-amino acid, the amino group of amino acid residues a) and b) being optionally mono-C₁₋₃ alkylated.

F is a group of formula —COOR_1 , $\text{—CH}_2\text{OR}_2$ or $\text{—CO—N} \begin{matrix} \text{R}_3 \\ \text{R}_4 \end{matrix}$

R₂ is hydrogen,

R₃ is hydrogen or C₁₋₃ alkyl,

R₄ is hydrogen or C₁₋₃ alkyl and

Y₁ and Y₂ together represent a direct bond, as well as the acid addition salts and complexes thereof.

The polypeptides of the invention may exist in salt form or in the form of complexes thereof. Acid addition salts may be formed with e.g. organic acids, polymeric acids and inorganic acids. Such acid

addition salt forms include e.g. the hydrochlorides and acetates. By complexes are to be understood compounds of known type, formed from compounds of formula I on addition of inorganic substances, e.g. inorganic salts or hydroxides such as Ca- and Zn-salts, and/or on addition of polymeric organic substances.

The present invention also provides a process for the production of the compounds according to the invention. These compounds may be produced by methods known in the art of peptide chemistry or by obvious chemical equivalents thereof, for example by a process comprising:

a) removing at least one protecting group from a protected polypeptide having the sequence indicated in formula I,

b) linking together by an amide bond two peptide units, each of which contains at least one amino acid or amino alcohol residue in protected or unprotected form, the peptide units being such that a protected or unprotected polypeptide having the sequence indicated in formula I, is obtained and, if necessary, carrying out process step a);

c) converting a functional group A or F of a protected or unprotected polypeptide having the sequence indicated in formula I, into another N- or C-terminal functional group, in particular another group A or F, and, if necessary carrying out process step a);

d) oxidising a straight chain polypeptide of formula I wherein Y_1 and Y_2 are each hydrogen to provide a polypeptide of formula I wherein Y_1 and Y_2 together are a direct bond, and when required converting a polypeptide thus obtained into a salt form or complex thereof.

The above process may for example be carried out analogously to the processes described in the accompanying examples. Insofar as the production of the starting materials is not particularly described, the compounds are known or may be produced and purified in accordance with methods known in the art.

The polypeptides of the invention as well as their pharmaceutically acceptable salts and complexes exhibit valuable pharmacological properties as indicated in animal tests. In particular they exhibit GH-secretion inhibiting activity as indicated e.g. by depression of serum GH-levels in the rat.

This test is carried out employing male rats under Nembutal narcosis. The test-substance is administered at varying logarithmically staggered doses employing at least 4 rats per dose. The rats are decapitated 15 minutes after administration, the blood is collected and the serum GH-level determined by radio-immuno-assay.

The said polypeptides, salts and complexes are accordingly indicated for use in the treatment of disorders with an aetiology comprising or associated with excess GH-secretion, e.g. in the treatment of diabetes mellitus and angiopathy as well as of acromegaly.

The said polypeptides, salts and complexes also inhibit gastric- and pancreatic secretion as indicated in standard animal tests e.g. in accordance with the method described in Scand. J. Gastroent. 6, 423 (1971) by Kouturek et al. for the pancreatic secretion and in accordance with the following method for the measurement of the gastric secretion:

Male rats (220—280 g) are kept in the laboratory in individual cages for several days (14 hours light/day) with 48 hours fasting (but free access to water) immediately prior to the experiment. At start of the experiment the pylorus is ligated under ether anaesthesia and 100 μ g/kg pentagastrin are injected s.c. to stimulate gastric secretion. The test compounds are injected simultaneously i.m.. 30 mins. later the animals are sacrificed, the volume of gastric juice measured and the acid concentration estimated (titration with thymol blue). The acid output is calculated and the percent inhibition estimated in relation to untreated controls.

The said polypeptides, salts and complexes are thus further indicated for use in the treatment of gastro-intestinal disorders, for example in the treatment of gastric ulcer, gastro-intestinal bleeding and acute pancreatitis. For the above uses an indicated daily dose is from about 0.002, e.g. from about 0.07, to about 20 mg polypeptide conveniently administered in divided doses 2 to 4 times a day in unit dosage or sustained release form. Suitable unit dosage forms contain from about 0.0005, e.g. from about 0.02, to about 10 mg, of a polypeptide in accordance with the invention or equivalent amount of a pharmaceutically acceptable salt or complex thereof, together with a solid or liquid pharmaceutical diluent or carrier therefor.

In accordance with the foregoing the present invention further provides: 1) a polypeptide in accordance with the invention, in particular a compound of formula I, or a pharmaceutically acceptable salt or complex thereof, for use as a pharmaceutical, (e.g. for use as a GH-secretion inhibitor in the treatment of disorders with an aetiology comprising or associated with excess GH-secretion, such as diabetes mellitus, angiopathy and acromegaly and for use as a gastric or pancreatic secretion inhibitor in the treatment of gastro-intestinal disorders such as gastric ulcer, gastro-intestinal bleeding and acute pancreatitis) as well as 2) pharmaceutical compositions comprising said polypeptides, salts and complexes.

In the following examples all temperatures are in °C and $[\alpha]_D^{20}$ - values are uncorrected. The following abbreviations are employed:

	AcOH	=	acetic acid
5	Asp-diol	=	the asparagine-diol residue $\text{HO}-\text{CH}_2-\text{CH}_2-\text{CH} \begin{array}{l} \text{NH-} \\ \text{CH}_2\text{OH} \end{array}$
	Boc	=	tert.-butoxycarbonyl
10	BTFA	=	boron-tris-trifluoroacetate
	DCCI	=	dicyclohexylcarbodiimide
15	DMF	=	N,N-dimethylformamide
	HOBT	=	N-hydroxybenzotriazole
	Hy	=	$-\text{NHNH}_2$
20	Leu-ol	=	the leucinol residue $\text{CH}_3-\text{CH}(\text{CH}_3)-\text{CH}_2-\text{CH} \begin{array}{l} \text{NH-} \\ \text{CH}_2\text{OH} \end{array}$
25	MBzl	=	p-methoxybenzyl
	Me	=	methyl
30	MeOH	=	methanol
	MePhe	=	-(N-methyl)-Phe-
35	NEt ₃	=	triethylamine
	ONP	=	4-Nitrophenoxy
	Phe(pF)	=	-p-F-Phe-
40	Phe-ol	=	the phenylalaninol residue $\text{C}_6\text{H}_5-\text{CH}_2-\text{CH} \begin{array}{l} \text{NH-} \\ \text{CH}_2\text{OH} \end{array}$
45	Ser-ol	=	the serinol residue $-\text{NH}-\text{CH}(\text{CH}_2\text{OH})_2$
	TFA	=	trifluoroacetic acid
50	THF	=	tetrahydrofuran
55	Thr-ol	=	the threoninol residue $\text{CH}_3-\text{CH}(\text{OH})-\text{CH} \begin{array}{l} \text{NH-} \\ \text{CH}_2\text{OH} \end{array}$
60	Z	=	benzyloxycarbonyl

Example 1A

H-(D)Phe-Cys-Phe-(D)Trp-Lys-Thr-Cys-Thr-ol

- 5 0.70 g Boc-(D)Phe-Cys(MBzl-Phe-(D)Trp-Lys(Boc)-Thr-Cys(MBzl)-Thr-ol and 6.0 ml of thioanisol are dissolved in 9 ml TFA at 0°. The solution is cooled to -10° and 12 ml ca. 2M BTFA in TFA are added with stirring. Stirring is continued for 1 1/2 hours at a temperature of -5° to -10° and 50 ml cold abs. MeOH (-70°) are added. After a further 5 min. 1 liter abs. ether and 5 ml ca. 5N HCl in ether are added with stirring. The precipitated product is filtered off, washed briefly with ether and the residue dissolved immediately in 2.5 l dioxan/H₂O (7:3). The pH is adjusted to pH 7—7.5 by the addition of dilute ammonia solution. The mixture is stirred in an open vessel at room temperature until it tests negative for —SH groups (e.g. Ellmann-Test). The pH is adjusted to 3—4 by addition of HCl and the obtained solution concentrated under vacuum, and the product lyophilised. The lyophilisate is dissolved in a small quantity 10% AcOH and purified by gel-chromatography (Bio-Rad P4) using 10% AcOH.
- 10 15 Fractions containing the desired product are collected and lyophilised, to yield the title compound: $[\alpha]_D^{20} -42^\circ$ (c=0.5 in 95%P AcOH).

The starting material may be obtained as follows:

- a) Z-Lys(Boc)-Thr-OMe
- 20 13.5 ml chloroformic acid isobutyl ester are added with stirring to 56 g Z-Lys(Boc)-OH in the form of the dicyclohexylamine salt dissolved in 1200 ml THF, precooled to -20°. Stirring is continued for a further 20 minutes at -15° and a cold solution (-20°) of 21 g HCl.H-Thr-OMe in 600 ml THF is then added. 17 ml triethylamine are added drop-wise at a temperature of -15°, the reaction mixture stirred for 18 hours at 0° and the product subjected to strong concentration under vacuum. Dilution is effected with ether/ethylacetate (1:1) and the precipitated dicyclohexylamine salt filtered off. The filtrate is washed out with H₂O, 2N citric acid, 10% KHCO₃ and H₂O. The organic phase is dried over Na₂SO₄ and evaporated to dryness, to yield the title compound as a resin: $[\alpha]_D^{20} = -14^\circ$ (c=1 in DMF).
- 25 b) Z-Phe-(D)Trp-OH
- 30 9.9 g Z-Phe-OH in 150 ml acetonitrile are cooled to -18° and 3.4 ml N-methyl-morpholine followed by 4.1 ml chloroformic acid isobutyl ester are added. The solution is stirred for 15 minutes at -15° and a cold solution of 6.2 g H-(D)Trp-OH in 30 ml 1N NaOH and 100 ml acetonitrile is added, and stirring continues for a further 18 hours at a temperature of -5° to 0°. The reaction mixture is concentrated under vacuum, diluted with water and extracted 3X with a small quantity of ether. The aqueous phase is adjusted to ca. pH 2 by the addition of 4N H₂SO₄ and the precipitate extracted with ether/acetic acid. The organic phase is washed with water and dried over Na₂SO₄. After evaporation under vacuum, the residue is crystallised from ether/petrolether to yield the title compound: m.p. 113°C, $[\alpha]_D^{20} = -5^\circ$ (c=0.9 in DMF).
- 35 c) Z-Phe-(D)Trp-Lys(Boc)-Thr-OMe
- 40 6.0 g Z-Lys(Boc)-Thr-OMe in 150 ml MeOH are hydrogenated in the presence of Pd/C. The product is filtered, washed with MeOH and the filtrate evaporated under vacuum. The residue is dissolved together with 4.8 g Z-Phe-(D)Trp-OH and 2.0 g HOBT in 50 ml DMF. The solution is cooled to -15° and 2.1 g DCCl in 15 ml DMF are added with stirring. The solution is stirred for a further 2 days at 0°, evaporated under vacuum, diluted with acetic acid/ether (1:1) and the precipitate dicyclohexylurea filtered off. The filtrate is washed with 2N citric acid, 10% KHCO₃ and H₂O. The organic phase is dried over Na₂SO₄ and evaporated. The residue is chromatographed over silica-gel with CH₂Cl₂/MeOH as eluant. The fractions containing the desired product are collected and evaporated under vacuum to yield the title compound: $[\alpha]_D^{20} = -7^\circ$ (c=0.9 in DMF).
- 45 d) H-Phe-(D)Trp-Lys(Boc)-Thr-OMe
- 50 1.2 g Z-Phe-(D)Trp-Lys(Boc)-Thr-OMe in 50 ml MeOH are hydrogenated in the presence of 10% Pd/C and the solution filtered and evaporated under vacuum to yield the title compound as a foam: $[\alpha]_D^{20} = -18^\circ$ (c=1 in MeOH).
- 55 e) Boc-(D)Phe-Cys(MBzl)-OH
- 60 8.4 ml NEt₃ followed by 8.3 ml chloroformic acid isobutyl ester are added to 16 g Boc-(D)Phe-OH in 300 ml THF precooled to -20°. The obtained mixture is stirred for 10 minutes at a temperature of -15° and a cold solution of H-Cys-(MBzl)-OH and 11.5 ml NEt₃ in 400 ml THF/H₂O (5:1) are subsequently added drop-wise. The reaction mixture is stirred for ca. 2 days at a temperature of 0°, concentrated under vacuum, diluted with 1.3 l H₂O and extracted with ether. The aqueous phase is adjusted to pH 2.0 by the addition of 4N H₂SO₄ and the precipitate extracted with ethyl acetate. The ethyl acetate extract is washed with water, dried over Na₂SO₄ and evaporated under vacuum. The residue is purified by chromatography over silica gel with ether/1% AcOH as eluant. The fractions

containing the desired product are combined and evaporated under vacuum to yield the title compound: $[\alpha]_D^{20} = -18^\circ$ ($c=1.1$ in DMF).

f) Boc-(D)Phe-Cys(MBzl)-Phe-(D)Trp-Lys(Boc)-Thr-OMe

0.30 g DCCl are added with stirring to 0.67 g Boc-(D)Phe-Cys(MBzl)-OH, 1.0 g H-Phe-(D)Trp-Lys(Boc)-Thr-OMe and 0.5 g HOBT dissolved in 30 ml DMF, pre-cooled to -20° . The reaction mixture is stirred for ca. 12 hours at a temperature of from -5° to 0° and for ca. 4 hours at room temperature. the precipitated dicyclohexyl urea is filtered off and the filtrate washed with 2N citric acid, 10% KHCO_3 and H_2O . The organic phase is dried over Na_2SO_4 purified and concentrated. The product is precipitated by the addition of ether, filtered and dried to yield the title compound: m.p. 160° , $[\alpha]_D^{20} = -30^\circ$ ($c=1$ in MeOH).

g) Boc-(D)Phe-Cys(MBzl)-Phe-(D)Trp-Lys(Boc)-Thr-NHNH₂

1.5 ml hydrazine hydrate are added to 0.9 g Boc-(D)Phe-Cys(MBzl)-Phe-(D)Trp-Lys(Boc)-Thr-OMe dissolved in 15 ml DMF. The solution is allowed to stand for 5 hours at room temperature and aqueous MeOH is added. The precipitate is filtered off, washed with MeOH/ H_2O (1:9) and dried to yield the title compound: m.p. 160° , $[\alpha]_D^{20} = -19^\circ$ ($c=1$ in DMF).

h) H-Cys(MBzl)-Thr-ol trifluoroacetate

A solution of 7.1 g Boc-Cys(MBzl)-Thr-ol and 5 ml thionanisole in 25 ml methylenchloride are added to 50 ml TFA and allowed to stand for 20 minutes at room temperature. The solution is diluted with ca. 1.5 l ether and the precipitate filtered off, washed with ether and dried to yield the title compound: $[\alpha]_D^{20} [\alpha]_D^{20} = 8.3^\circ$ ($c=1.1$ in 95% AcOH).

i) Boc-Cys(MBzl)-Thr-ol

2.1 ml N-methylmorpholine are added with stirring to 6.3 g Boc-Cys(MBzl)-OH in 50 ml THF pre-cooled to -20° , followed by the drop-wise addition at a temperature of -15° of 2.4 ml chloroformic acid isobutyl ester. After stirring for 5 minutes at a temperature of -15° a cold solution (-10°) comprising 3.3 g H-Thr-ol hydrochloride and 4.1 ml N-methylmorpholine in 30 ml DMF are added. The mixture is stirred for 2 hours at 0° and for a further 2 hours at room temperature. 50 ml 10% KHCO_3 are added, the reaction mixture concentrated under vacuum, diluted with ethyl acetate and washed 3 x with 2N citric acid, 3 x with 10% KHCO_3 and subsequently with 30% NaCl solution. The organic phase is dried over Na_2SO_4 and evaporated under vacuum to yield the title compound as an oil: $[\alpha]_D^{20} = -31^\circ$ ($c=1.3$ in DMF).

j) Boc-(D)Phe-Cys(MBzl)-Phe-(D)Trp-Lys(Boc)-Thr-Cys(MBzl)-Thr-ol

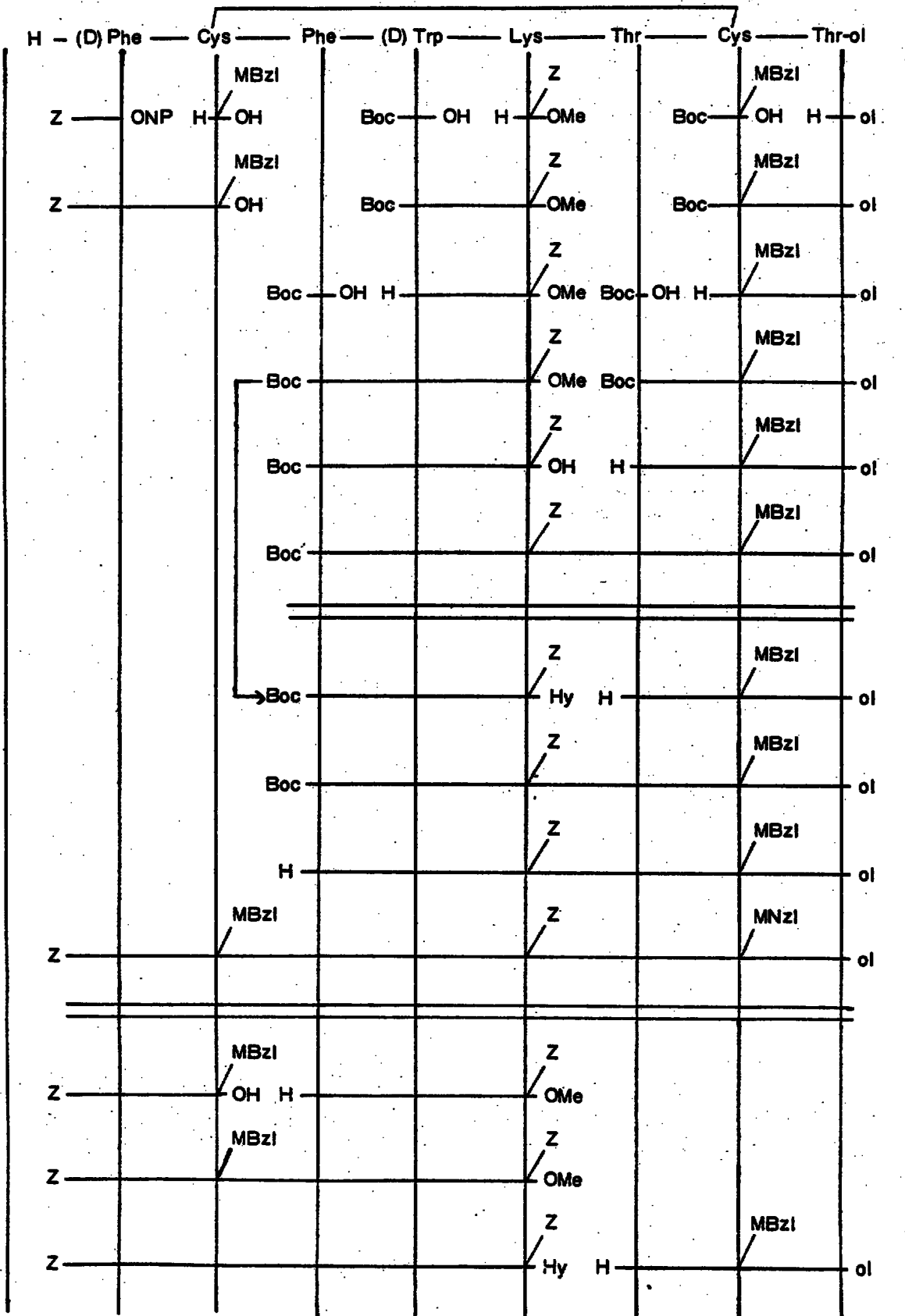
0.7 ml ca. 5N HCl in ether are added with stirring to 0.82 g Boc-(D)Phe-Cys(MBzl)-Phe-(D)Trp-Lys(Boc)-Thr-NH-NH₂ in 30 ml DMF pre-cooled to -20° , followed by, in toto, 0.8 ml, 10% tert.butylinitrite in DMF. The mixture is stirred for 15 minutes at a temperature of -20° to -15° . 0.56 ml NEt_3 are added at a temperature of -25° , followed by a solution of 0.51 g H-Cys (MBzl)-Thr-ol hydrochloride in 3 ml DMF, pre-cooled to -15° , and the whole is stirred for 20 hours at -5° to 0° and for a further 3 hours at room temperature. The reaction mixture is diluted with ca. 100 ml methanol and the product precipitated by addition of ca. 40 ml H_2O . The precipitate is filtered off, washed with aqueous MeOH and dried to yield the title compound: decomposition at 170° , $[\alpha]_D^{20} = -20^\circ$ ($c=1$ in DMF).

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EXAMPLE 1B

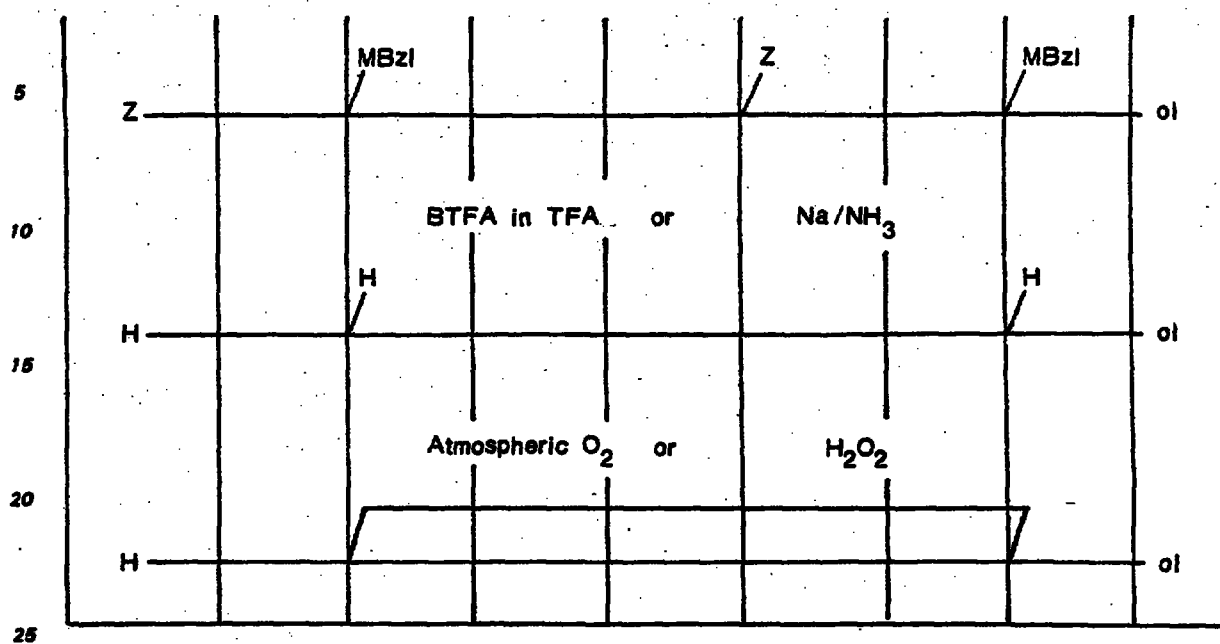
The product of Example 1A may also be prepared in accordance with the following synthetic sequences:



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Example 1B (continued)

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The following compounds may be produced analogously to the above described processes (all compounds as acetate unless otherwise indicated):

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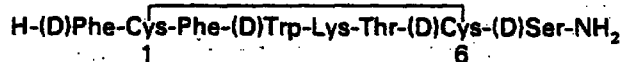
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Example No.	A	E	F'	$[\alpha]_D^{20}$ in 95% AcOH
2	H-(D) Phe-	-Thr-	-(D) Ser-NH ₂	-48° (c = 1.0)
3	H-(D) Phe-	-Thr-	-NH ₂	-35° (c = 1.0)
4	H-(D) Phe-	-Thr-	-Phe-ol	-41° (c = 1.0)
5	H-(D) Phe-	-Thr-	-Leu-ol	-41° (c = 1.0)
6	H-(D) Phe-	-Thr-	-Asp-diol	-42° (c = 1.0)
7	H-Nle-	-Thr-	-(S) Ser-NH ₂	-42° (c = 1.0)
8	H-(D) Phe-	-(D) Ala-	-(D) Ser-NH ₂	-23° (c = 0.9)
9	H-Asn-	-Thr-	-(D) Ser-NH ₂	-28° (c = 0.5)
10	H-(D) MePhe-	-Thr-	-Thr-ol	-63° (c = 1.0)
11	H-Nle-Asn-	-Thr-	-Thr-ol	-40° (c = 1.0)
12	H-(D) Phe-	-(D) Ala-	-Thr-ol	-9° (c = 1.0)
13	H-(D) Phe-	-Thr-	-Ser-ol	-43° (c = 1.1)
14	H-(D) Phe-	-(D) Thr-	-Thr-ol	-73° (c = 1.0)
15	 -(CH ₂) ₂ -CO-	-The-	-Thr-ol	-25° (c = 0.9)
16	H-(D) Phe(pF)-	-Thr-	-Thr-ol	-41° (c = 0.5)

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Example 17



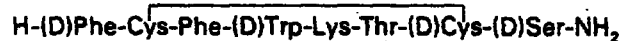
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HCl salt: $[\alpha]_D^{20}$ in 95% AcOH = -85° (c=0.4).

Example 18



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$[\alpha]_D^{20}$ in 95% AcOH = -102° (c=1.0).

Example 19



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$[\alpha]_D^{20}$ in 95% A-OH = -30° (c=1.0).

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$$[\alpha]_D^{20} \text{ in } 95\% \text{ AcOH} = -42^\circ \text{ (c=1.0)}$$
$$[\alpha]_D^{20} \text{ in } 95\% \text{ AcOH} = -35^\circ \text{ (c=0.5).}$$

$$[\alpha]_D^{20} \text{ in } 95\% \text{ AcOH} = -42^\circ \text{ (c=1.0)}$$
$$[\alpha]_D^{20} \text{ in } 95\% \text{ AcOH} = -35^\circ (c=0.5).$$

1 4 5 6 7 8 9 0



0 029 579

R_1 and R_2 have the meanings given above,
 R_3 is hydrogen or C_{1-3} alkyl and
 R_7 is hydrogen, C_{1-3} alkyl, phenyl or C_{7-10} phenylalkyl, the group $—CH(R_5)—X$ having the D- or L-configuration, and

Y_1 and Y_2 are each hydrogen or together represent a direct bond, whereby the residues in the 1- and 6-position each independently have the L- or D-configuration, with the proviso that D- and/or L-cysteine residues are present at the 1- and 6-positions only, as well as the salt forms and complexes thereof.

2. A polypeptide according to claim 1, wherein

A is C_{1-3} alkyl, C_{7-10} phenylalkyl or a group of formula $RCO—$,

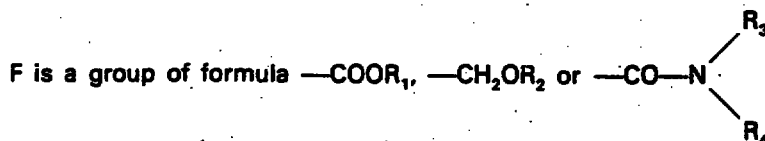
A' is hydrogen,

R is hydrogen, C_{1-3} alkyl, phenyl or C_{7-10} phenylalkyl or

RCO is a) an L- or D-phenylalanine residue optionally mono-ring-substituted by F, Cl, Br, NO_2 , NH_2 , OH, C_{1-3} alkyl or C_{1-3} alkoxy, or

b) the residue of a natural α -amino acid other than defined under a) above or of a corresponding D-amino acid,

the amino group of amino acid residues a) and b) being optionally mono- C_{1-3} alkylated,



R_2 is hydrogen,

R_3 is hydrogen or C_{1-3} alkyl,

R_7 is hydrogen or C_{1-3} alkyl and

Y_1 and Y_2 together represent a direct bond, and B, C, D, E, R_1 , R_4 , R_5 , R_6 and X have the meanings given in claim 1,

as well as the acid addition salts and complexes thereof.

3. A polypeptide according to claim 1 of formula



as well as the acid addition salt forms and complexes thereof.

4. A polypeptide according to claim 1 of formula



as well as the acid addition salt forms and complexes thereof.

5. A polypeptide according to claim 1 of formula



wherein

a) A is H-(D)Phe-, E is -Thr- and F' is selected from the group consisting of -(D)Ser- NH_2 , $-NH_2$, -Phe-ol, -Leu-ol, -Asp-diol and -Ser-ol;

b) A is selected from the group consisting of H-Nle-Asn-,  $—(CH_2)_2—CO—$ and

H-(D)Phe(pF)-, E is -Thr- and F' is -Thr-ol;

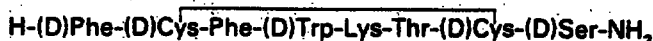
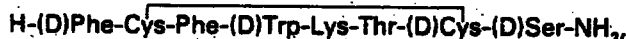
c) A is H-(D)Phe-, E is selected from the group consisting of -(D)Ala- and -(D)Thr- and F' is -Thr-ol;

d) A is H-(D)Phe-, E is -(D)Ala- and F' is -(D)Ser- NH_2 ; or

e) A is H-Asn-, E is -Thr- and F' is -(D)Ser- NH_2 ,

as well as the acid addition salt forms and complexes thereof.

6. A polypeptide according to claim 1 of formula



as well as the acid addition salt forms and complexes thereof.



b) linking together by an amide bond two peptide units, each of which contains at least one amino acid or amino alcohol residue in protected or unprotected form, the peptide units being such that a protected or unprotected polypeptide having the sequence of formula I of claim 1 is obtained and, if necessary, carrying out process step a);

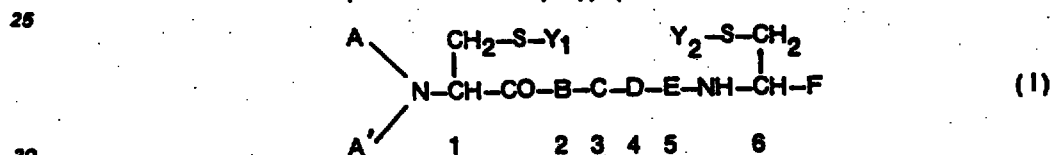
10 d) oxidising a straight chain polypeptide of formula I wherein Y₁ and Y₂ are each hydrogen to provide a polypeptide of formula I of claim 1, wherein Y₁ and Y₂ together are a direct bond, and when required converting a polypeptide thus obtained into a salt form or complex thereof.

8. A polypeptide according to any one of claims 1 to 6 or a pharmaceutically acceptable salt form or complex thereof for use as a pharmaceutical.

15 9. A polypeptide according to any one of claims 1 to 6 or a pharmaceutically acceptable salt form or complex thereof for use in the treatment of disorders with an aetiology comprising or associated with excess GH-secretion, or for use as a gastric- or pancreatic secretion inhibitor.

10. A pharmaceutical composition comprising a polypeptide according to any one of claims 1 to 6 or a pharmaceutically acceptable salt form or complex thereof together with a pharmaceutically acceptable diluent or carrier therefor.

1. Process for the production of a polypeptide of formula I,



A is C₁₋₁₂ alkyl, C₇₋₁₀ phenylalkyl or a group of formula RCO—, whereby

i) R is hydrogen, C₁₋₁₁ alkyl, phenyl or C₇₋₁₀ phenylalkyl, or,

ii) RCO— is

a) an L- or D-phenylalanine residue optionally ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl and/or C₁₋₃ alkoxy

b) the residue of a natural α -amino acid other than defined under a) above or of a corresponding D-amino acid, or

40 c) a dipeptide residue in which the individual amino acid residues are the same or different and are selected from those defined under a) and/or b) above, the α -amino group of amino acid residues a) and b) and the N-terminal amino group of dipeptide residues c) being optionally mono- or di-C₁₋₁₂ alkylated,

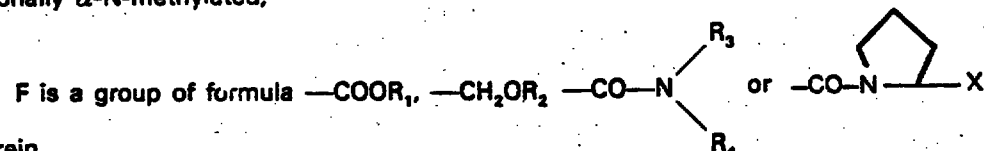
A' is hydrogen or, when A is C₁₋₁₂ alkyl or C₇₋₁₀ phenylalkyl, also C₁₋₁₂ alkyl or C₇₋₁₀ phenylalkyl,

B is -Phe- optionally ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl and/or C₁₋₃ alkoxy,

C is -(L)- or -(D)Trp- optionally α -N-methylated and optionally benzene-ring-substituted by F, Cl, Br, NO₂, NH₂, OH, C₁₋₃ alkyl and/or C₁₋₃ alkoxy.

D is -Lys- optionally α -N-methylated and optionally ϵ -N-C₁₋₃ alkylated,

E is the residue of a natural α -amino acid or of a corresponding D-amino acid, said residue being
50 optionally α -N-methylated,



55 wherein

R_1 is hydrogen or C_{1-3} alkyl,

R_2 is hydrogen or the residue of a physiologically acceptable, physiologically hydrolysable ester,

R₁ is hydrogen or the residue of a physiologically absorbable ester;
R₂ is hydrogen, C₁₋₃ alkyl, phenyl or C₇₋₁₀ phenylalkyl,

60 R_5 is hydrogen, $-(CH_2)_2-OH$ or $-(CH_2)_3-OH$, or represents the substituent attaching to the α -carbon atom of a natural α -amino acid and



X is a group of formula —COOR_1 , $\text{—CH}_2\text{OR}_2$ or $\text{—CO—N} \begin{matrix} \text{R}_6 \\ \text{R}_7 \end{matrix}$ wherein

R_1 and R_2 have the meanings given above,

R_6 is hydrogen or C_{1-3} alkyl and

R_7 is hydrogen, C_{1-3} alkyl, phenyl or C_{7-10} phenylalkyl, the group $\text{—CH(R}_5\text{)—X}$ having the D- or L-configuration, and

Y_1 and Y_2 are each hydrogen or together represent a direct bond, whereby the residues in the 1- and 6-position each independently have the L- or D-configuration, with the proviso that D- and/or L-cysteine residues are present at the 1- and 6-positions only, as well as the salt forms and complexes thereof, which process comprises

a) removing at least one protecting group from a protected polypeptide having the sequence of formula I;

b) linking together by an amide bond two peptide units, each of which contains at least one amino acid or amino alcohol residue in protected or unprotected form, the peptide units being such that a protected or unprotected polypeptide having the sequence of formula I is obtained and, if necessary, carrying out process step a);

c) converting a functional group A or F of a protected or unprotected polypeptide having the sequence of formula I, into another group A or F, and, if necessary, carrying out process step a);

d) oxidising a straight chain polypeptide of formula I, wherein Y_1 and Y_2 are each hydrogen to provide a polypeptide of formula I, wherein Y_1 and Y_2 together are a direct bond, and when required converting a polypeptide thus obtained into a salt form or complex thereof.

2. Process according to claim 1, wherein, in the polypeptide of formula I,

A is C_{1-3} alkyl, C_{7-10} phenylalkyl or a group of formula RCO— ,

A' is hydrogen,

R is hydrogen, C_{1-3} alkyl, phenyl or C_{7-10} phenylalkyl or

RCO is a) an L- or D-phenylalanine residue optionally mono-ring-substituted by F, Cl, Br, NO_2 , NH_2 , OH, C_{1-3} alkyl or C_{1-3} alkoxy, or

b) the residue of a natural α -amino acid other than defined under a) above or of a corresponding D-amino acid,

the amino group of amino acid residues a) and b) being optionally mono- C_{1-3} alkylated,

F is a group of formula —COOR_1 , $\text{—CH}_2\text{OR}_2$ or $\text{—CO—N} \begin{matrix} \text{R}_3 \\ \text{R}_4 \end{matrix}$

R_2 is hydrogen,

R_3 is hydrogen or C_{1-3} alkyl,

R_7 is hydrogen or C_{1-3} alkyl and

Y_1 and Y_2 together represent a direct bond, and B, C, D, E, R, R_4 , R_5 , R_6 and X have the meanings given in claim 1, as well as the acid addition salts and complexes thereof and wherein the initially obtained polypeptide is, when required, converted into an acid addition salt form or complex thereof.

3. Process according to claim 1, wherein, in the polypeptide of formula I, A is H-(D)Phe-, A' is hydrogen, B is -Phe-, C is -(D)Trp-, D is -Lys-, E is -Thr-, F is -Leu-ol, Y_1 and Y_2 together represent a direct bond and the residues at the 1- and 6-position each have the L-configuration.

4. Process according to claim 1, wherein, in the polypeptide of formula I, A is H-(D)Phe-, A' is hydrogen, B is -Phe-, C is -(D)Trp-, D is -Lys-, E is -Thr-, F is -Thr-ol, Y_1 and Y_2 together represent a direct bond and the residues at the 1- and 6-position each have the L-configuration.

5. Process according to claim 1, wherein, in the polypeptide of formula I, A' is hydrogen, B is -Phe-, C is -(D)Trp-, D is -Lys-, Y_1 and Y_2 together represent a direct bond, the residues at the 1- and 6-position each have the L-configuration and

a) A is H-(D)Phe-, E is -Thr- and F' is selected from the group consisting of -(D)Ser- NH_2 , —NH_2 , -Phe-ol, -Leu-ol, -Asp-diol and -Ser-ol;

b) A is selected from the group consisting of H-Nle-Asn,  $\text{—(CH}_2\text{)}_2\text{—CO—}$ and

H-(D)Phe(pF)-, E is -Thr- and F' is -Thr-ol;

c) A is H-(D)Phe-, E is selected from the group consisting of -(D)Ala- and -(D)Thr- and F' is -Thr-ol;

d) A is H-(D)Phe-, E is -(D)Ala- and F' is -(D)Ser- NH_2 ; or

e) is H-Asn-, E is -Thr- and F' is -(D)Ser- NH_2 ,

as well as the acid addition salt forms and complexes thereof.



6. Process according to claim 1, wherein, in the polypeptide of formula I, A is H-(D)Phe-, A' is hydrogen, C is -(D)Trp-, D is -Lys-, E is -Thr- and

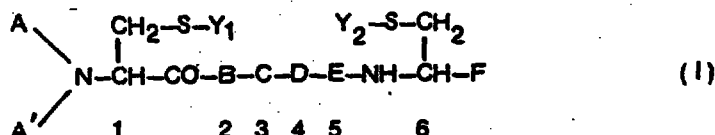
a) B is -Phe-, F is -(D)Ser-NH₂, Y₁ and Y₂ together represent a direct bond and (i) the residue at the 1-position has the L-configuration and the residue at the 6-position has the D-configuration, or (ii) the residue at the 6-position has the L-configuration and the residue at the 1-position has the D-configuration, or (iii) the residues at the 1- and 6-positions both have the D-configuration;

b) B is -Phe(pF)-, F is -Thr-ol, Y₁ and Y₂ together represent a direct bond and the residues at the 1- and 6-positions both have the L-configuration; or

c) B is -Phe-, F is -Thr-ol, Y₁ and Y₂ are each hydrogen and the residues at the 1- and 6-positions both have the L-configuration.

Revendications pour les Etats contractants: BE CH DE FR GB IT LI LU NL SE

1. Un polypeptide de formule I,



dans laquelle

A signifie un groupe alkyle en C₁-C₁₂, phénylalkyle en C₇-C₁₀ ou un groupe de formule RCO—, dans laquelle

i) R signifie l'hydrogène, un groupe alkyle en C₁-C₁₁, phényle ou phénylalkyle en C₇-C₁₀, ou

ii) RCO— signifie

a) un reste L- ou D-phénylalanine éventuellement substitué dans le noyau par F, Cl, Br, NO₂, NH₂, OH, alkyle en C₁-C₃ et/ou alcoxy en C₁-C₃,

b) le reste d'un α-amino-acide naturel autre que défini sous a) ci-dessus, ou d'un D-amino-acide correspondant, ou

c) un reste dipeptidique dans lequel les restes de chaque amino-acide sont identiques ou différents et sont choisis parmi ceux définis sous a) et/ou b) ci-dessus, le groupe α-amino des restes d'amino-acides a) et b) et le groupe amino N-terminal des restes dipeptidiques c) étant éventuellement mono- ou dialkylés par des restes alkyle en C₁-C₁₂,

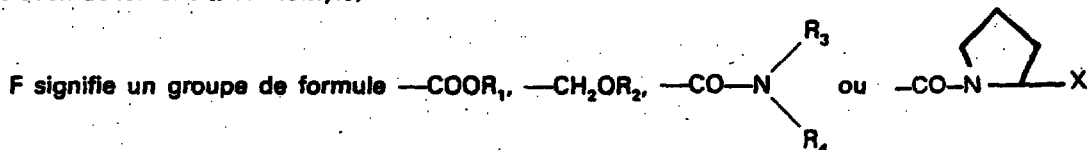
A' signifie l'hydrogène ou, lorsque A signifie un groupe alkyle en C₁-C₁₂ ou phénylalkyle en C₇-C₁₀, également un groupe alkyle en C₁-C₁₂ ou phénylalkyle en C₇-C₁₀,

B signifie -Phe- éventuellement substitué dans le noyau par F, Cl, Br, NO₂, NH₂, OH, alkyle en C₁-C₃ et/ou alcoxy en C₁-C₃,

C signifie -(L)- ou (D)Trp- éventuellement α-N-méthylé et éventuellement substitué dans le noyau benzénique par F, Cl, Br, NO₂, NH₂, OH, alkyle en C₁-C₃ et/ou alcoxy en C₁-C₃,

D signifie -Lys- éventuellement α-N-méthylé et éventuellement ε-N-alkylé par un reste alkyle en C₁-C₃,

E signifie le reste d'un α-amino-acide naturel ou d'un D-amino-acide correspondant, ledit reste étant éventuellement α-N-méthylé,



dans lesquelles

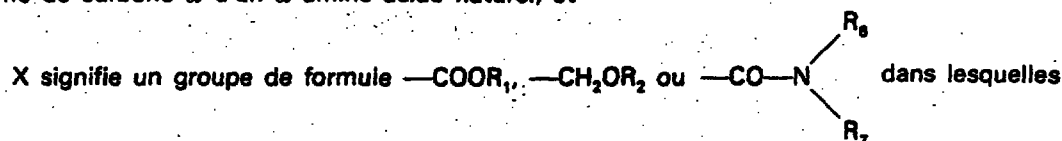
R₁ signifie l'hydrogène ou un groupe alkyle en C₁-C₃,

R₂ signifie l'hydrogène ou le reste d'un ester physiologiquement hydrolysable, physiologiquement acceptable,

R₃ signifie l'hydrogène, un groupe alkyle en C₁-C₃, phényle ou phénylalkyle en C₇-C₁₀,

R₄ signifie l'hydrogène, un groupe alkyle en C₁-C₃ ou, lorsque R₃ signifie l'hydrogène ou un groupe méthyle, également un groupe de formule $\text{—CH(R}_5\text{)—X}$,

R₅ signifie l'hydrogène, $\text{—(CH}_2\text{)}_2\text{—OH}$ ou $\text{—(CH}_2\text{)}_3\text{—OH}$, ou représente le substituant attaché à l'atome de carbone α d'un α-amino-acide naturel, et



R₁ et R₂ ont les significations données ci-dessus,



R_6 signifie l'hydrogène ou un groupe alkyle en C_1-C_3 , et
 R_7 signifie l'hydrogène, un groupe alkyle en C_1-C_3 , phényle ou phénylalkyle en C_7-C_{10} , le groupe
 $-CH(R_8)-X$ ayant la configuration D ou L, et

Y_1 et Y_2 signifient chacun l'hydrogène ou représentent ensemble une liaison directe,
 5 les restes situés en position 1 et 6 ayant chacun, indépendamment l'un de l'autre, la configuration L ou D, avec la condition que des restes D- et/ou L-cystéine soient uniquement présents aux positions 1 et 6, ainsi que les sels et les complexes de ce polypeptide.

2. Un polypeptide selon la revendication 1, caractérisé en ce que

A signifie un groupe alkyle en C_1-C_3 , phénylalkyle en C_7-C_{10} ou un groupe de formule $RCO-$,

A' signifie l'hydrogène,

R signifie l'hydrogène, un groupe alkyle en C_1-C_3 , phényle ou phénylalkyle en C_7-C_{10} ou
 RCO signifie

a) un reste L- ou D-phénylalanine éventuellement monosubstitué dans le noyau par F, Cl, Br, NO_2 , NH_2 , OH, alkyle en C_1-C_3 ou alcoxy en C_1-C_3 , ou

b) le reste d'un α -amino-acide naturel autre que défini sous a) ci-dessus ou d'un D-amino-acide correspondant,

le groupe amino des restes d'amino-acides a) et b) étant éventuellement monoalkylé par un reste alkyle en C_1-C_3 ,

F signifie un groupe de formule $-COOR_1$, $-CH_2OR_2$ ou $-CO-N$



R_2 signifie l'hydrogène,

R_3 signifie l'hydrogène ou un groupe alkyle en C_1-C_3 ,

R_4 signifie l'hydrogène ou un groupe alkyle en C_1-C_3 et

Y_1 et Y_2 représentent ensemble une liaison directe, et B, C, D, E, R_1 , R_4 , R_5 , R_6 et X ont les significations données à la revendication 1, ainsi que les sels d'addition d'acides et les complexes de ce polypeptide.

3. Un polypeptide selon la revendication 1, de formule



ainsi que les sels d'addition d'acides et les complexes de ce polypeptide.

4. Un polypeptide selon la revendication 1, de formule



ainsi que les sels d'addition d'acides et les complexes de ce polypeptide.

5. Un polypeptide selon la revendication 1, de formule



dans laquelle

a) A signifie H-(D)-Phe, E signifie -Thr- et F' est choisi dans le groupe constitué par -(D)Ser- NH_2 , - NH_2 , -Phe-ol, -Leu-ol, -Asp-di-ol et -Ser-ol;

b) A est choisi dans le groupe constitué par H-Nle-Asn-,  $(CH_2)_2-CO-$ et

H-(D)Phe(pF)-, E signifie -Thr- et F' signifie -Thr-ol;

c) A signifie H-(D)-Phe-, E est choisi dans le groupe constitué par -(D)-Ala- et -(D)Thr- et F' signifie -Thr-ol;

d) A signifie H-(D)Phe-, E signifie -(D)Ala- et F' signifie -(D)Ser- NH_2 ; ou

e) A signifie H-Asn-, E signifie -Thr- et F' signifie -(D)Ser- NH_2 , ainsi que les sels d'addition d'acides et les complexes de ce polypeptide.

6. Un polypeptide selon la revendication 1, de formule



ainsi que les sels d'addition d'acides et les complexes de ce polypeptide.

7. Procédé de préparation d'un composé selon la revendication 1, lequel procédé comprend

a) l'élimination d'au moins un groupe protecteur à partir d'un polypeptide protégé ayant la séquence de la formule I comme défini à la revendication 1,

5 b) le couplage par une liaison amide de deux unités peptidiques, lesquelles contiennent chacune au moins un reste d'acide-amino ou d'acide-amino-alcool sous forme protégée ou non protégée, les unités peptidiques étant telles qu'on obtient un polypeptide protégé ou non-protégé ayant la séquence de la formule I selon la revendication 1, et, si nécessaire, la mise en oeuvre du procédé de l'étape a);

c) la transformation d'un groupe fonctionnel A ou F d'un polypeptide protégé ou non protégé 10 ayant la séquence de la formule I selon la revendication 1, en un autre groupe A ou F, et, si nécessaire, la mise en oeuvre du procédé de l'étape a);

d) l'oxydation d'un polypeptide à chaîne linéaire de formule I selon la revendication 1, dans laquelle Y_1 et Y_2 sont chacun l'hydrogène, pour donner un polypeptide de formule I selon la revendication 1, dans laquelle Y_1 et Y_2 forment ensemble une liaison directe, et, si nécessaire, la trans- 15 formation d'un polypeptide ainsi obtenu en un sel ou complexe de ce polypeptide.

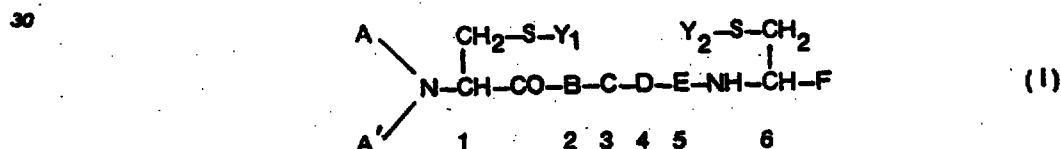
8. Un polypeptide selon l'une quelconque des revendications 1 à 6 ou un sel ou un complexe pharmaceutiquement acceptables de ce polypeptide, pour l'utilisation comme médicament.

9. Un polypeptide selon l'une quelconque des revendications 1 à 6 ou un sel ou complexe pharmaceutiquement acceptables de ce polypeptide, pour l'utilisation dans le traitement des troubles 20 avec une étiologie comprenant ou associée à un excès de sécrétion de l'hormone de croissance, ou pour l'utilisation comme inhibiteur de la sécrétion gastrique ou pancréatique.

10. Une composition pharmaceutique comprenant un polypeptide selon l'une quelconque des revendications 1 à 6 ou un sel ou complexe pharmaceutiquement acceptables de ce polypeptide, en association avec un diluant ou véhicule pharmaceutiquement acceptables.

Revendications pour l'Etat contractant: AT

1. Procédé de préparation d'un polypeptide de formule I,



dans laquelle

A signifie un groupe alkyle en $C_1\text{---}C_{12}$, phénylalkyle en $C_7\text{---}C_{10}$ ou un groupe de formule RCO--- , dans laquelle

i) R signifie l'hydrogène, un groupe alkyle en $C_1\text{---}C_{11}$, phényle ou phénylalkyle en $C_7\text{---}C_{10}$, ou

40 ii) RCO--- signifie

a) un reste L- ou D-phénylalanine éventuellement substitué dans le noyau par F, Cl, Br, NO_2 , NH_2 , OH, alkyle en $C_1\text{---}C_3$ et/ou alcoxy en $C_1\text{---}C_3$,

b) le reste d'un α -amino-acide naturel autre que défini sous a) ci-dessus, ou d'un D-amino-acide correspondant, ou

45 c) un reste dipeptidique dans lequel les restes de chaque amino-acide sont identiques ou différents et sont choisis parmi ceux définis sous a) et/ou b) ci-dessus, le groupe α -amino des restes d'amino-acides a) et b) et le groupe amino N-terminal des restes dipeptidiques c) étant éventuellement mono- ou dialkylés par des restes alkyle en $C_1\text{---}C_{12}$,

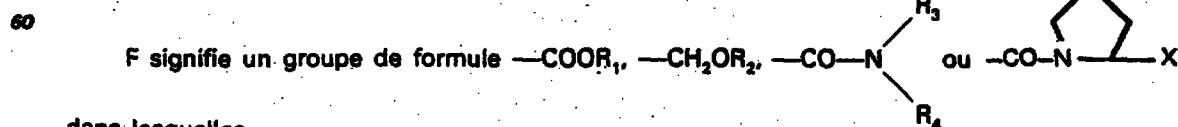
A' signifie l'hydrogène ou, lorsque A signifie un groupe alkyle en $C_1\text{---}C_{12}$ ou phénylalkyle en 50 $C_7\text{---}C_{10}$, également un groupe alkyle en $C_1\text{---}C_{12}$ ou phénylalkyle en $C_7\text{---}C_{10}$.

B signifie -Phe- éventuellement substitué dans le noyau par F, Cl, Br, NO_2 , NH_2 , OH, alkyle en $C_1\text{---}C_3$ et/ou alcoxy en $C_1\text{---}C_3$,

C signifie -(L)- ou (D)Trp- éventuellement α -N-méthylé et éventuellement substitué dans le noyau benzénique par F, Cl, Br, NO_2 , NH_2 , OH, alkyle en $C_1\text{---}C_3$ et/ou alcoxy en $C_1\text{---}C_3$,

55 D signifie -Lys- éventuellement α -N-méthylé et éventuellement ϵ -N-alkylé par un reste alkyle en $C_1\text{---}C_3$,

E signifie le reste d'un α -amino-acide naturel ou d'un D-amino-acide correspondant, ledit reste étant éventuellement α -N-méthylé,



dans lesquelles

65 R_1 signifie l'hydrogène ou un groupe alkyle en $C_1\text{---}C_3$.



R_2 signifie l'hydrogène ou le reste d'un ester physiologiquement hydrolysable, physiologiquement acceptable,

R_3 signifie l'hydrogène, un groupe alkyle en C_1-C_3 , phényle ou phénylalkyle en C_7-C_{10} ,

R_4 signifie l'hydrogène, un groupe alkyle en C_1-C_3 ou, lorsque R_3 signifie l'hydrogène ou un groupe méthyle, également un groupe de formule $-CH(R_5)-X$,

R_5 signifie l'hydrogène, $-(CH_2)_2-OH$ ou $-(CH_2)_3-OH$, ou représente le substituant attaché à l'atome de carbone α d'un α -amino-acide naturel, et

X signifie un groupe de formule $-COOR_1$, $-CH_2OR_2$ ou $-CO-N$ dans lesquelles



R_1 et R_2 ont les significations données ci-dessus,

R_6 signifie l'hydrogène ou un groupe alkyle en C_1-C_3 , et

R_7 signifie l'hydrogène, un groupe alkyle en C_1-C_3 , phényle ou phénylalkyle en C_7-C_{10} , le groupe $-CH(R_5)-X$ ayant la configuration D ou L, et

Y_1 et Y_2 signifient chacun l'hydrogène ou représentent ensemble une liaison directe, les restes situés en position 1 et 6 ayant chacun, indépendamment l'un de l'autre, la configuration L ou D, avec la condition que des restes D- et/ou L-cystéine soient uniquement présents aux positions 1 et 6, ainsi que les sels et les complexes de ce polypeptide, lequel procédé comprend

a) l'élimination d'au moins un groupe protecteur à partir d'un polypeptide protégé ayant la séquence de la formule I;

b) la couplage par une liaison amide de deux unités peptidiques, lesquelles contiennent chacune au moins un reste d'acide-amino ou d'acide-amino-alcool sous forme protégée ou non protégée, les unités peptidiques étant telles qu'on obtient un polypeptide protégé ou non-protégé ayant la séquence de la formule I, et, si nécessaire, la mise en oeuvre du procédé de l'étape a);

c) la transformation d'un groupe fonctionnel A ou F d'un polypeptide protégé ou non protégé ayant la séquence de la formule I, en un autre groupe A ou F, et, si nécessaire, la mise en oeuvre du procédé de l'étape a);

d) l'oxydation d'un polypeptide à chaîne linéaire de formule I, dans laquelle Y_1 et Y_2 sont chacun l'hydrogène, pour donner un polypeptide de formule I, dans laquelle Y_1 et Y_2 forment ensemble une liaison directe, et, si nécessaire, la transformation d'un polypeptide ainsi obtenu en un sel ou complexe de ce polypeptide.

2. Procédé selon la revendication 1, caractérisé en ce que, dans le polypeptide de formule I,

A signifie un groupe alkyle en C_1-C_3 , phénylalkyle en C_7-C_{10} ou un groupe de formule $RCO-$,

A' signifie l'hydrogène,

R signifie l'hydrogène, un groupe alkyle en C_1-C_3 , phényle ou phénylalkyle en C_7-C_{10} ou RCO signifie

a) un reste L- ou D-phénylalanine éventuellement monosubstitué dans le noyau par F, Cl, Br, NO_2 , NH_2 , OH, alkyle en C_1-C_3 ou alcoxy en C_1-C_3 , ou

b) le reste d'un α -amino-acide naturel autre que défini sous a) ci-dessus ou d'un D-amino-acide correspondant,

le groupe amino des restes d'amino-acides a) et b) étant éventuellement monoalkylé par un reste alkyle en C_1-C_3 .

F signifie un groupe de formule $-COOR_1$, $-CH_2OR_2$ ou $-CO-N$



R_2 signifie l'hydrogène,

R_3 signifie l'hydrogène ou un groupe alkyle en C_1-C_3 ,

R_4 signifie l'hydrogène ou un groupe alkyle en C_1-C_3 et

Y_1 et Y_2 représentent ensemble une liaison directe, et B, C, D, E, R_1 , R_4 , R_5 , R_6 et X ont les significations données à la revendication 1, ainsi que les sels d'addition d'acides et les complexes de ce polypeptide, et dans lequel le polypeptide obtenu initialement est, si nécessaire, transformé en un sel d'addition d'acide ou un complexe de ce polypeptide.

3. Procédé selon la revendication 1, caractérisé en ce que, dans le polypeptide de formule I, A signifie H-(D)Phe-, A' signifie l'hydrogène, B signifie -Phe-, C signifie -(D)Trp-, D signifie -Lys-, E signifie -Thr-, F signifie -Leu-ol, Y_1 et Y_2 représentent ensemble une liaison directe et les restes situés en position 1 et 6 ont chacun la configuration L.

4. Procédé selon la revendication 1, caractérisé en ce que, dans le polypeptide de formule I, A signifie H-(D)Phe-, A' signifie l'hydrogène, B signifie -Phe-, C signifie -(D)Trp-, D signifie -Lys-, E signifie -Thr-, F signifie -Thr-ol, Y_1 et Y_2 représentent ensemble une liaison directe et les restes situés en position 1 et 6 ont chacun la configuration L.

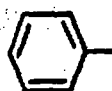
5. Procédé selon la revendication 1, caractérisé en ce que, dans la polypeptide de formule I, A'



signifie l'hydrogène, B signifie -Phe-, C signifie -(D)Trp-, D signifie -Lys-, Y₁ et Y₂ représentent ensemble une liaison directe, les restes situés en position 1 et 6 ont chacun la configuration L et

a) A signifie H-(D)-Phe-, E signifie -Thr- et F' est choisi dans le groupe constitué par -(D)Ser-NH₂, -NH₂, -Phe-ol-, -Leu-ol-, -Asp-diol et -Ser-ol;

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b) A est choisi dans le groupe constitué par H-Nle-Asn-,  (CH₂)₂-CO- et

H-(D)Phe(pF)-, E signifie -Thr- et F' signifie -Thr-ol;

c) A signifie H-(D)-Phe-, E est choisi dans le groupe constitué par -(D)-Ala- et -(D)Thr- et F' signifie -Thr-ol;

d) A signifie H-(D)Phe-, E signifie -(D)Ala- et F' signifie -(D)Ser-NH₂; ou

e) A signifie H-Asn-, E signifie -Thr- et F' signifie -(D)Ser-NH₂, ainsi que les sels d'addition d'acides et les complexes de ce polypeptide.

6. Procédé selon la revendication 1, caractérisé en ce que, dans le polypeptide de formule I, A

signifie H-(D)Phe-, A' signifie l'hydrogène, C signifie -(D)Trp-, D signifie -Lys-, E signifie -Thr- et

a) B signifie -Phe-, F signifie -(D)Ser-NH₂, Y₁ et Y₂ représentent ensemble une liaison directe, et

(i) le reste situé en position 1 a la configuration L et le reste situé en position 6 a la configuration

D, ou bien

(ii) le reste situé en position 6 a la configuration L et le reste situé en position 1 a la configuration

D, ou bien

(iii) les restes situés aux positions 1 et 6 ont tous les deux la configuration D;

b) B signifie -Phe(pF)-, F signifie -Thr-ol-, Y₁ et Y₂ représentent ensemble une liaison directe et les

restes situés aux positions 1 et 6 ont tous les deux la configuration L; ou

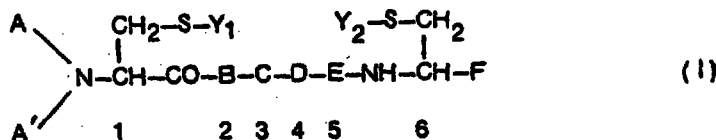
c) B signifie -Phe-, F signifie -Thr-ol-, Y₁ et Y₂ signifient chacun l'hydrogène et les restes situés aux

positions 1 et 6 ont tous les deux la configuration L.

Patentansprüche für die Vertragsstaaten: BE CH DE FR GB IT LI LU NL SE

1. Ein polypeptid der Formel I

30



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worin

A für C₁₋₁₂-Alkyl, C₇₋₁₀-Phenylalkyl oder für ein Gruppe der Formel RCO— steht, wobei

1) R Wasserstoff, C₁₋₁₁-Alkyl, Phenyl oder C₇₋₁₀-Phenylalkyl, oder

40

2) RCO—

a) einen gegebenenfalls im Ring durch F, Cl, Br, NO₂, NH₂, OH, C₁₋₃-Alkyl und/oder C₁₋₃-Alkoxy substituierten L- oder D-Phenylalaninrest,

b) den Rest einer anderen als oben unter a) definierten natürlichen α-Aminosäure oder einer entsprechenden D-Aminosäure, oder

45

c) einen Dipeptidrest, worin die einzelnen Aminosäurereste gleich oder verschieden sind und aus den oben unter a) und/oder b) definierten Resten ausgewählt werden, bedeuten, wobei die α-Aminogruppe der Aminosäurereste a) und b) und die N-terminale Aminogruppe des Dipeptidrestes c) gegebenfalls durch C₁₋₁₂-Alkyl mono- oder di-alkyliert sind,

A' für Wasserstoff oder, falls A C₁₋₁₂-Alkyl oder C₇₋₁₀-Phenylalkyl bedeutet, auch für C₁₋₁₂-Alkyl

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oder C₇₋₁₀-Phenylalkyl steht,

B für gegebenenfalls im Ring durch F, Cl, Br, NO₂, NH₂, OH, C₁₋₃-Alkyl und/oder C₁₋₃-Alkoxy substituiertes -Phe- steht,

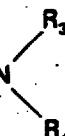
C für gegebenenfalls α-N-methyliertes und gegebenenfalls im Benzolring durch F, Cl, Br, NO₂, NH₂, OH, C₁₋₃-Alkyl und/oder C₁₋₃-Alkoxy substituiertes (L)- oder (D)-Trp steht,

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D für gegebenenfalls α-N-methyliertes und gegebenenfalls ε-N-C₁₋₃-alkyliertes -Lys- steht,

E für den gegebenenfalls α-N-methylierten Rest einer natürlichen α-Aminosäure oder einer entsprechenden D-Aminosäure steht,

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F für eine Gruppe der Formel  oder  X steht,

worin

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R, Wasserstoff oder C₁₋₃-Alkyl





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R_2 Wasserstoff oder den Rest eines physiologisch verträglichen, physiologisch hydrolysierbaren Esters,

R_3 Wasserstoff, C_{1-3} -Alkyl, Phenyl oder C_{7-10} -Phenylalkyl

R_4 Wasserstoff, C_{1-3} -Alkyl oder, falls R_3 für Wasserstoff oder Methyl steht, auch eine Gruppe der

5 Formel $-\text{CH}(R_5)-\text{X}$

R_5 Wasserstoff, $-(\text{CH}_2)_2-\text{OH}$ oder $-(\text{CH}_2)_3-\text{OH}$, oder den am α -Kohlenstoffatom befindlichen Substituent einer natürlichen α -Aminosäure und

10 X eine Gruppe der Formel $-\text{COOR}_1$, $-\text{CH}_2\text{OR}_2$ oder $-\text{CO}-\text{N} \begin{matrix} \nearrow R_6 \\ \searrow R_7 \end{matrix}$, worin

R_1 und R_2 die oben erwähnten Bedeutungen haben,

R_6 für Wasserstoff oder C_{1-3} -Alkyl und

15 R_7 für Wasserstoff, C_{1-3} -Alkyl, Phenyl oder C_{7-10} -Phenylalkyl stehen, bedeuten, wobei die Gruppe $-\text{CH}(R_5)-\text{X}$ die D- oder L-Konfiguration hat, und

Y_1 und Y_2 beide für Wasserstoff oder zusammen für eine direkte Bindung stehen, wobei die Reste in den Stellungen 1 und 6 unabhängig voneinander die L- oder D-Konfiguration haben, mit der Massgabe, dass D- und/oder L-Cysteinreste nur in den Stellungen 1 und 6 vorkommen, sowie seine Salz-

20 forme und Komplexe.

2. Ein Polypeptid gemäss Anspruch 1, worin

A für C_{1-3} -Alkyl, C_{7-10} -Phenylalkyl oder eine Gruppe der Formel $\text{RCO}-$,

A' für Wasserstoff

R für Wasserstoff, C_{1-3} -Alkyl, Phenyl oder C_{7-10} -Phenylalkyl oder

25 RCO für

a) einen gegebenenfalls im Ring durch F, Cl, Br, NO_2 , NH_2 , OH, C_{1-3} -Alkyl oder C_{1-3} -Alkoxy monosubstituierten L- oder D-Phenylalaninrest, oder

b) den Rest einer anderen als unter a) definierten natürlichen α -Aminosäure oder einen entsprechenden D-Aminosäure, wobei die Aminogruppe der Aminosäurereste a) und b) gegebenenfalls

30 durch C_{1-3} -Alkyl monoalkyliert ist,

F für eine Gruppe der Formel $-\text{COOR}_1$, $-\text{CH}_2\text{OR}_2$ oder $-\text{CO}-\text{N} \begin{matrix} \nearrow R_3 \\ \searrow R_4 \end{matrix}$

35 R_2 für Wasserstoff

R_3 für Wasserstoff oder C_{1-3} -Alkyl

R_7 für Wasserstoff oder C_{1-3} -Alkyl und

Y_1 und Y_2 zusammen für eine direkte Bindung stehen, und B, C, D, E, R_1 , R_4 , R_5 , R_6 und X die im

40 Anspruch 1 angegebenen Bedeutungen haben, sowie seine Säureadditionssalze und Komplexe.

3. Ein Polypeptid gemäss Anspruch 1 der Formel



45 sowie seine Säureadditionssalze und Komplexe.

4. Ein Polypeptid gemäss Anspruch 1 der Formel



50 sowie seine Säureadditionssalze und Komplexe.

5. Ein Polypeptid gemäss Anspruch 1 der Formel



worin

55 a) A für H-(D)Phe und E für -Thr- stehen und F' aus der Gruppe bestehend aus -(D)Ser-NH₂, -NH₂, -Phe-ol, -Leu-ol, -Asp-di-ol und -Ser-ol ausgewählt wird,

b) A aus der Gruppe bestehend aus H-Nle-Asn-,  $(\text{CH}_2)_2-\text{CO}$ und

H-(D)Phe(pF)-ausgewählt wird, E für -Thr- und F' für -Thr-ol stehen;

60 c) A für H-(D)Phe- steht, E aus der Gruppe bestehend aus -(D)Ala- und -(D)Thr- ausgewählt wird und F' für -Thr-ol steht;

d) A für H-(D)Phe-, E für -(D)Ala- und F' für -(D)Ser-NH₂ stehen; oder

e) A für H-Asn-, E für -Thr- und F' für -(D)Ser-NH₂ stehen, sowie seine Säureadditionssalze und Komplexe.

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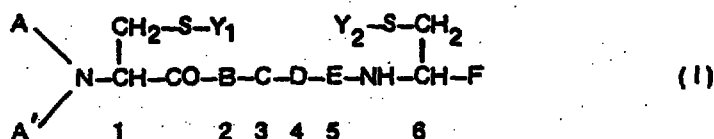


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C für gegebenenfalls α -N-methyliertes und gegebenenfalls im Benzolring durch F, Cl, Br, NO₂, NH₂, OH, C₁₋₃-Alkyl und/oder C₁₋₃-Alkoxy substituiertes (L)- oder (D)-Trp steht,

D für gegebenenfalls α -N-methyliertes und gegebenenfalls ϵ -N-C₁₋₃-alkyliertes -Lys- steht,

E für den gegebenenfalls α -N-methylierten Rest einer natürlichen α -Aminosäure oder einer entsprechenden D-Aminosäure steht,

F für eine Gruppe der Formel —COOR_1 , CH_2OR_2 , $\text{CON} \begin{matrix} \text{R}_3 \\ \text{R}_4 \end{matrix}$ oder $\text{—CO—N} \begin{matrix} \text{ } \\ \text{ } \end{matrix} \text{X}$ steht,

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worin

R₁ Wasserstoff oder C₁₋₃-Alkyl

R₂ Wasserstoff oder den Rest eines physiologisch verträglichen, physiologisch hydrolysierbaren Esters,

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R₃ Wasserstoff, C₁₋₃-Alkyl, Phenyl oder C₇₋₁₀-Phenylalkyl

R₄ Wasserstoff, C₁₋₃-Alkyl oder, falls R₃ für Wasserstoff oder Methyl steht, auch eine Gruppe der Formel $\text{—CH(R}_5\text{)—X}$

R₅ Wasserstoff, $\text{—(CH}_2\text{)}_2\text{—OH}$ oder $\text{—(CH}_2\text{)}_3\text{—OH}$, oder den am α -Kohlenstoffatom befindlichen Substituent einer natürlichen α -Aminosäure und

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X eine Gruppe der Formel —COOR_1 , $\text{—CH}_2\text{OR}_2$ oder $\text{—CO—N} \begin{matrix} \text{R}_6 \\ \text{R}_7 \end{matrix}$ worin

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R₁ und R₂ die oben erwähnten Bedeutungen haben,

R₆ für Wasserstoff oder C₁₋₃-Alkyl und

R₇ für Wasserstoff, C₁₋₃-Alkyl, Phenyl oder C₇₋₁₀-Phenylalkyl stehen, bedeuten, wobei die Gruppe $\text{—CH(R}_5\text{)—X}$ die D- oder L-Konfiguration hat, und

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Y₁ und Y₂ beide für Wasserstoff oder zusammen für eine direkte Bindung stehen, wobei die Reste in den Stellungen 1 und 6 unabhängig voneinander die L- oder D-Konfiguration haben, mit der Massgabe, dass D- und/oder L-Cysteinreste nur in den Stellungen 1 und 6 vorkommen, sowie seiner Salzform und Komplexe, dadurch gekennzeichnet, dass man

a) mindestens eine Schutzgruppe aus einem geschützten Polypeptid mit der in der Formel I angegebenen Sequenz entfernt,

b) zwei Peptideinheiten, von denen jede mindestens einen Aminosäure- oder Aminoalkoholrest in geschützter oder ungeschützter Form enthält, durch eine Amidbindung miteinander verknüpft, wobei die Peptideinheiten so gewählt werden, dass ein geschütztes oder ungeschütztes Polypeptid mit der Sequenz der Formel I erhalten wird und, wenn notwendig, die Verfahrensstufe a) ausführt;

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c) eine funktionelle Gruppe A oder F eines geschützten oder ungeschützten Polypeptids mit der Sequenz der Formel I in eine andere Gruppe A oder F überführt und, wenn notwendig, die Verfahrensstufe a) ausführt;

d) ein geradkettiges Polypeptid der Formel I, worin Y₁ und Y₂ beide Wasserstoff bedeuten, zu einem Polypeptid der Formel I, worin Y₁ und Y₂ zusammen für eine direkte Bindung stehen, Oxidiert, und gewünschtenfalls ein so erhaltenes Polypeptid in eine Salzform oder Komplex davon überführt.

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2. Verfahren gemäss Anspruch 1, dadurch gekennzeichnet, dass im Polypeptid der Formel I

A für C₁₋₃-Alkyl, C₇₋₁₀-Phenylalkyl oder eine Gruppe der Formel RCO—,

A' für Wasserstoff

R für Wasserstoff, C₁₋₃-Alkyl, Phenyl oder C₇₋₁₀-Phenylalkyl oder

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RCO für

a) einen gegebenenfalls im Ring durch F, Cl, Br, NO₂, NH₂, OH, C₁₋₃-Alkyl oder C₁₋₃-Alkoxy monosubstituierten L- oder D-Phenylalaninrest, oder

b) den Rest einer anderen als unter a) definierten natürlichen α -Aminosäure oder einen entsprechenden D-Aminosäure, wobei die Aminogruppe der Aminosäurereste a) und b) gegebenenfalls durch C₁₋₃-Alkyl monoalkyliert ist,

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F für eine Gruppe der Formel —COOR_1 , $\text{—CH}_2\text{OR}_2$ oder $\text{—CO—N} \begin{matrix} \text{R}_3 \\ \text{R}_4 \end{matrix}$

60

R₂ für Wasserstoff

R₃ für Wasserstoff oder C₁₋₃-Alkyl

R₇ für Wasserstoff oder C₁₋₃-Alkyl und

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Y₁ und Y₂ zusammen für eine direkte Bindung stehen, und B, C, D, E, R₁, R₄, R₅, R₆ und X die im



Anspruch 1 angegebenen Bedeutungen haben, sowie seine Säureadditionssalze und Komplexe, und dass das anfänglich erhaltene Polypeptid gewünschtenfalls in ein Säureadditionssalz oder Komplex davon überführt wird.

3. Verfahren gemäss Anspruch 1, dadurch gekennzeichnet, dass im Polypeptid der Formel I A für H-(D)Phe-, A' für Wasserstoff, B für -Phe-, C für -(D)Trp-, D für -Lys-, E für -Thr-, F für -Leu-ol und Y₁ und Y₂ zusammen für eine direkte Bindung stehen und die Reste in den Stellungen 1 und 6 beide in L-Konfiguration haben.

4. Verfahren gemäss Anspruch 1, dadurch gekennzeichnet, dass im Polypeptid der Formel I A für H-(D)Phe-, A' für Wasserstoff, B für -Phe-, C für -(D)Trp-, D für -Lys-, E für -Thr-, F für -Thr-ol und Y₁ und Y₂ zusammen für eine direkte Bindung stehen und die Reste in den Stellungen 1 und 6 beide die L-Konfiguration haben.

5. Verfahren gemäss Anspruch 1, dadurch gekennzeichnet, dass im Polypeptid der Formel I A' für Wasserstoff, B für -Phe-, C für -(D)Trp-, D für -Lys- und Y₁ und Y₂ zusammen für eine direkte Bindung stehen, die Reste in den Stellungen 1 und 6 beide die L-Konfiguration haben und

a) A für H-(D)Phe und E für -Thr- stehen und F' aus der Gruppe bestehend aus -(D)-Ser-NH₂, -NH₂, -Phe-ol, -Leu-ol, -Asp-diol und -Ser-ol ausgewählt wird,

b) A aus der Gruppe bestehend aus H-Nle-Asn-, , (CH₂)₂-CO und

H-(D)Phe(pF)-ausgewählt wird, E für -Thr- und F' für -Thr-ol stehen;

c) A für H-(D)Phe- steht, E aus der Gruppe bestehend aus -(D)Ala- und -(D)Thr- ausgewählt wird und F' für -Thr-ol steht;

d) A für H-(D)Phe-, E für -(D)Ala- und F' für -(D)Ser-NH₂ stehen; oder

e) A für H-Asn-, E für -Thr- und F' für -(D)Ser-NH₂ stehen, sowie seine Säureadditionssalze und Komplexe.

6. Verfahren gemäss Anspruch 1, dadurch gekennzeichnet, dass im Polypeptid der Formel I A für H-(D)Phe-, A' für Wasserstoff, C für -(D)Trp-, D für -Lys-, E für -Thr- und

a) B für -Phe-, F für -(D)Ser-NH₂, Y₁ und Y₂ zusammen für eine direkte Bindung stehen und

1) der Rest in Stellung 1 die L-Konfiguration und der Rest in Stellung 6 die D-Konfiguration oder

2) der Rest in Stellung 1 die D-Konfiguration und der Rest in Stellung 6 die L-Konfiguration oder

3) die Reste in den Stellungen 1 und 6 beide die D-Konfiguration haben;

b) B für -Phe(pF)-, F für -Thr-ol, Y₁ und Y₂ zusammen für eine direkte Bindung stehen und die Reste in den Stellungen 1 und 6 beide die L-Konfiguration haben; oder

c) B für -Phe-, F für -Thr-ol, Y₁ und Y₂ beide für Wasserstoff stehen und die Reste in den Stellungen 1 und 6 beide die L-Konfiguration haben.

