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(54) Benævnelse: **Matrix-fikserede beta-hårnåleformede peptidomimetika med proteasehæmmende aktivitet**

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Description

[0001] The present invention provides template-fixed beta-hairpin peptidomimetics incorporating a template-fixed chain of 11 amino acid residues, which, depending on their position in the chain, are as defined hereinbelow. These template-fixed beta-hairpin peptidomimetics are useful as inhibitors of protease enzymes. They are especially valuable as inhibitors of various serine proteases, such as human cathepsin G, elastase, or tryptase. In addition, the present invention provides an efficient process, by which these compounds can, if desired, be made in library-format.

[0002] The beta-hairpin peptidomimetics of the invention show improved efficacy, oral bioavailability, improved half-life and, most importantly, a high selectivity ratio among different serine proteases, which depends on the proper choice of certain types of amino acid residues and their position in said chain. In addition, these beta-hairpin peptidomimetics show a low hemolysis on red blood cells and low cytotoxicity.

[0003] Compounds with structures, which show some similarity to the structures of the beta-hairpin peptidomimetics of the invention, are known from WO-A-2002070547, which describes compounds with antimicrobial and anticancer activity; from WO-A-2003054000, which describes compounds useful as inhibitors of protease enzymes; from WO-A-2004018503 and WO-A-2004033489, which describe compounds with antimicrobial activity; and from WO-A-2004096838 and WO-A-2004096839, which describe compounds useful as antagonists of the chemokine receptor CXCR4.

[0004] Inhibitors of proteases are emerging with promising therapeutic uses in the treatment of diseases, such as cancers (R. P. Beckett et al., Drug Disc. Today 1996, 1, 16-26; L. L. Johnson et al., Curr. Opin. Chem. Biol. 1998, 2; 466-71; D. Leung et al., J. Med. Chem. 2000, 43, 305-341; T. Rockway, Expert Opin. Ther. Patents 2003, 13, 773-786), parasitic, fungal, and viral infections [e. g. schistosomiasis (M. M. Becker et al., J. Biol. Chem. 1995, 270, 24496-501*); C. albicans* (C. Abad-Zapetero et al., Protein Sci. 1996, 5, 640-52), HIV (A. Wlodawer et al., Annu. Rev. Biochem. 1993, 62, 543-85; P. L. Darke et al., Adv. Pharmacol. 1994, 5, 399-454), hepatitis (J. L. Kim et al., Cell 1996, 87, 343-55; R. A. Love et al., Cell 1996, 87, 331-342), herpes (W. Gibson et al., Drug Des. Discov. 1997, 15, 39-47)], and inflammatory, immunological, respiratory (P. R. Bernstein et al., Prog. Med. Chem. 1994, 31, 59-120; T. E. Hugli, Trends Biotechnol. 1996, 14, 409-12), cardiovascular (M. T. Stubbs et al., Thromb. Res. 1993, 69, 1-58; H. Fukami et al., Current Pharmaceutical Design 1998, 4, 439-453), and neurodegenerative defects, including Alzheimer's disease (R. Vassar et al., Science 1999, 286, 735-41), angiogenesis (M. Kaatinen et al., Atherosklerosis 1996, 123, 1-2, 123-131), and multiple sclerosis (M. Z. Ibrahim et al., J. Neuroimmunol. 1996, 70, 131-138).

[0005] As most proteases bind their substrates in extended or beta-strand conformations, good inhibitors must be able to mimick such a conformation. Beta-hairpin mimetics are thus ideally suited to lock peptide sequences in an extended conformation.

[0006] Among proteases, serine proteases constitute important therapeutic targets. Serine proteases are classified by their substrate specificity, particularly by the type of residue

found at P1, as either trypsin-like (positively charged residues Lys/Arg preferred at P1), elastase-like (small hydrophobic residues Ala/Val at P1), or chymotrypsin-like (large hydrophobic residues Phe/Tyr/Leu at P1). Serine proteases, for which protease-inhibitor X-ray crystal data are available on the PDB data base (PDB: www.rcsb.org/pdb), include trypsin, alpha-chymotrypsin, gamma-chymotrypsin, human neutrophil elastase, thrombin, subtilisin, human cytomegalovirus, proteinase A, achromobacter, human cathepsin G, glutamic acid-specific protease, carbopeptidase D, blood coagulation factor VIIa, porcine factor 1XA, mesentericopeptidase, HCV protease, and thermitase. Other serine proteases, which are of therapeutic interest, include tryptase, complement convertase, and hepatitis C-NS3 protease. Inhibitors of thrombin (e. g. J. L. Metha et al., J. Cardiovasc. Pharmacol. 1998, 31, 345-51; C. Lila et al., Synth. Comm. 1998, 28, 4419-29), and factor Xa (e. g. J. P. Vacca, Annu. Rep. Med. Chem. 1998, 33, 81-90) are in clinical evaluation as anti-thrombotics, inhibitors of elastase (J. R. Williams et al., Am. Rev. Respir. Dis. 1991, 144, 875-83) are in clinical trials for emphysema and other pulmonary diseases, whereas tryptase inhibitors are currently in phase II clinical trials for asthma (C. Seife, Science 1997, 277, 1602-3), urokinase inhibitors for breast cancer, and chymase inhibitors for heart related diseases. Finally, cathepsin G and elastase are intimately involved in the modulation of activities of cytokines and their receptors. Particularly at sites of inflammation, high concentration of cathepsin G, elastase and proteinase 3 are released from infiltrating polymorphonuclear cells in close temporal correlation to elevated levels of inflammatory cytokines, strongly indicating that these proteases are involved in the control of cytokine bioactivity and availability (U. Bank et al., J. Leukoc. Biol. 2001, 69, 177-90). Thus, inhibitors of elastase and cathepsin G constitute valuable targets for novel drug candidates, particularly for chronic obstructive pulmonary disease (H. Ohbayashi, Epert Opin. Investig. Drugs 2002, 11, 965-980).

[0007] Of the many occurring proteinaceous serine protease inhibitors, one is a 14 amino acids cyclic peptide from sunflower seeds, termed sunflower trypsin inhibitor (**SFTI-1**; S. Luckett et al., J. Mol. Biol. 1999, 290, 525-533; Y.-Q. Long et al., Biorg. & Med. Chem. Lett. 2001, 11, 2515-2519), which shows both sequence and conformational similarity with the trypsin-reactive loop of the Bowman-Birk family of serine protease inhibitors. The inhibitor adopts a beta-hairpin conformation, when bound to the active site of bovine beta-trypsin. **SFTI-1** inhibited beta-trypsin (K_i < 0.1nM), cathepsin G (K_i ~ 0.15nM), elastase (K_i ~ 105microM), chymotrypsin (K_i ~ 7.4microM), and thrombin (K_i ~ 136mM).

[0008] We illustrate here an approach to inhibitor design, which involves transplanting the beta-hairpin loop from the naturally occurring peptide onto a hairpin-inducing template. Based on the well-defined 3D-structure of the beta-hairpin mimetics, libraries of compounds can be designed, which ultimately can lead to novel inhibitors showing different specificity profiles towards several classes of proteases.

[0009] Template-bound hairpin mimetic peptides have been described in the literature (D. Obrecht et al., Adv. Med. Chem. 1999, 4, 1-68; J. A. Robinson, Syn. Lett. 2000, 4, 429-441), and serine protease-inhibiting template-fixed peptidomimetics and methods for their synthesis have been described in the International Patent Application WO 2003/054000 A1 and in A. Descours et al., ChemBioChem 2002, 3, 318-323, but the previously disclosed molecules do not exhibit high selectivity and particularly high potency. However, the ability to generate beta-hairpin peptidomimetics using combinatorial and parallel

synthesis methods has now been established (L. Jiang et al., Helv. Chim. Acta 2000, 83, 3097-3112).

[0010] These methods allow the synthesis and screening of large hairpin mimetic libraries, which in turn considerably facilitates structure-activity studies, and hence the discovery of new molecules with highly potent and selective serine protease inhibitory activity, oral bioavailability, low hemolytic activity to human red blood cells and low cytotoxicity.

[0011] The beta-hairpin peptidomimetics of the present invention, which can be present in free form or in pharmaceutically acceptable salt form, are defined hereinafter, e. g. in claim 1, and can be prepared in accordance with the present invention by a process defined hereinafter, e. g. in claim 40 or claim 41.

[0012] The peptidomimetics of the present invention can also be present in enantiomeric form, which can be prepared by a process defined hereinafter, e. g. in claim 42.

[0013] The dipeptide residue mentioned in claim 1 (hereinafter designated as "template") has an N-terminus and a C-terminus oriented in space in such a way, that the distance between those two may lie between 4.0 and 5.5 A. The undecapeptide chain Z mentioned in claim 1 is linked to the C-terminus and the N-terminus of the template via its corresponding N- and C-termini, so that the template and the chain Z form a cyclic structure, such as that depicted in the formula I shown in claim 1. In a case as here, where the distance between the N- and C-termini of the template lies between 4.0 and 5.5 A, the template will induce the H-bond network necessary for the formation of a beta-hairpin conformation in the chain Z. Thus, the template and the chain Z form a beta-hairpin mimetic.

[0014] The beta-hairpin conformation is highly relevant for the serine protease inhibitory activity of the beta-hairpin mimetics of the present invention. The beta-hairpin stabilizing conformational properties of the template play a key role not only for the selective inhibitory activity, but also for the synthesis process defined hereinabove, as incorporation of the template at the beginning or near the middle of the linear protected peptide precursors enhances cyclization yields significantly.

[0015] Side chains of L-amino acids at opposite positions of the beta-strand region can form an interstrand linkage. The most widely known linkage is the disulfide bridge formed by cysteines positioned at opposite positions of the beta-strand. Various methods are known to form disulfide linkages, including those described by J. P. Tam et al., Synthesis 1979, 955-957; Stewart et al., Solid Phase Peptide Synthesis, 2d Ed., Pierce Chemical Company, Ill., 1984; Ahmed et al., J. Biol. Chem. 1975, 250, 8477-8482; and Pennington et al., Peptides, pages 164-166, Giralt and Andreu, Eds., ESCOM Leiden, The Netherlands, 1990. Most advantageously, for the scope of the present invention, disulfide linkages can be prepared using acetamidomethyl (Acm) protective groups for cysteine. A further well-established interstrand linkage consists in linking a lysine residue with a glutamic acid residue, located at opposite beta-strand positions, by means of an amide bond formation. A preferred protective group for the side chain amino group of lysine is the allyloxycarbonyl (Alloc) group, and the side chain carboxy group of glutamic acid is preferably protected as allyl ester.

[0016] Positions for an interstrand linkage are P2 and P10, taken together. Such interstrand linkages are known to stabilize beta-hairpin conformations and, thus, constitute an important structural element for the design of beta-hairpin mimetics.

[0017] Hereinafter, those amino acids are listed, the residues of which make up the chain Z, the abbreviations and codes being those generally used.

[0018] Preferred amino acid residues in the chain Z are those derived from the following natural alpha-amino acids:

Three letter code	Name	One letter code
Ala	L-Alanine	A
Arg	L-Arginine	R
Asn	L-Asparagine	N
Asp	L-Aspartic acid	D
Cys	L-Cysteine	C
Gln	L-Glutamine	Q
Glu	L-Glutamic acid	E
His	L-Histidine	H
Ile	L-Isoleucine	I
Leu	L-Leucine	L
Lys	L-Lysine	K
Met	L-Methionine	M
Phe	L-Phenylalanine	F
Pro	L-Proline	P
Ser	L-Serine	S
Thr	L-Threonine	T
Trp	L-Tryptophan	W
Tyr	L-Tyrosine	Y

[0019] Other amino acid residues in the chain Z are those derived from the following amino acids:

Abbreviation	Name
Abu	Gamma-Aminobutyric acid (GABA)
AlloThr	Allo-Threonine
4Am Phe	L-para-Aminophenylalanine
Aoc	(2S)-Aminooctanoic acid

Abbreviation	Name
C_5al	L-3-Cyclopentylalanine
Cha	L-Cyclohexylalanine
2Cl-Phe	L-2-Chlorophenylalanine
Dpr	2,3-Diaminopropionic acid
hPhe	L-Homo-phenylalanine
1-Nal	L-1-Naphthylalanine
2-Nal	L-2-Naphthylalanine
Nle	L-Norleucine
OctG	L-Octylglycine
Phg	L-Phenylglycine
Pro(4NHCOPhe)	(2*S*)-4-benzamidino-pyrrolidine-2-carboxylic acid
t-BuA	L-t-Butylalanine

[0020] The positions P1 to P11 of each of the 11 amino acid residues in the chain Z are unequivocally defined as follows: P1 represents the first amino acid residue in the chain Z, that is coupled with its N-terminus to the C-terminus of the template, and P11 represents the last amino acid residue in the chain Z, that is coupled with its C-terminus to the N-terminus of the template.

[0021] For inhibitors of cathepsin G, the amino acid residues in the chain Z are preferably those defined in any one of claims 2 and 5 to 10.

[0022] For inhibitors of elastase, the amino acid residues in the chain Z are preferably those defined in any one of claims 3 and 11 to 22.

[0023] For inhibitors of tryptase, the amino acid residues in the chain Z are preferably those defined in any one of claims 4 and 23 to 25.

[0024] The amino acid building blocks, which make up the template, are those derived from the following amino acids, the abbreviations being those generally used (the amino acid ^{L}**Pro** is identical with the amino acid Pro):

Abbreviation	Name
^{D}Pro	D-Proline
^{L}Pro	L-Proline

[0025] Particularly preferred beta-hairpin peptidomimetics of the invention include those described in examples 5, 19, 20, 22, 23, 38, 39 and 40 as inhibitors of cathepsin G; in

examples 91, 153, 154, 155, 156, 157, 158, 159, 160 and 161 as inhibitors of elastase; and in examples 193, 194 and 195 as inhibitors of tryptase.

[0026] The processes of the invention can advantageously be carried out as parallel array syntheses to yield libraries of template-fixed beta-hairpin peptidomimetics of the invention. Such parallel syntheses allow to obtain arrays of numerous (normally 24 to 192, typically 96) compounds of the invention in high yields and defined purities, minimizing the formation of dimeric and polymeric by-products. The proper choice of the functionalized solid-support (i. e. solid support plus linker molecule), template and site of cyclization play thereby key roles.

[0027] The functionalized solid support is conveniently derived from polystyrene crosslinked with, preferably 1 to 5% of, divinylbenzene; polystyrene coated with polyethyleneglycol spacers (Tentagel®); and polyacrylamide resins (see also D. Obrecht et al., "Solid-Supported Combinatorial and Parallel Synthesis of Small-Molecular-Weight Compound Libraries", Tetrahedron Organic Chemistry Series, 17, Pergamon, Elsevier Science, 1998**).**

[0028] The solid support is functionalized by means of a linker, i. e. a bifunctional spacer molecule, which contains at the one end an anchoring group for attachment to the solid support and on the other end a selectively cleavable functional group used for the subsequent chemical transformations and cleavage procedures. For the purposes of the present invention, two types of linkers are used:

> Type 1 linkers are designed to release the amide group under acidic conditions (H. Rink, Tetrahedron Lett. 1987, 28, 3783-3790). Linkers of this kind form amides of the carboxyl group of the amino acid; examples of resins functionalized by such linker structures include 4-[(((2,4-dimethoxyphenyl)Fmoc-aminomethyl)-phenoxyacetamido)aminomethyl] PS resin, 4-[(((2,4-dimethoxyphenyl)Fmoc-aminomethyl)phenoxyacetamido) aminomethyl]-4-methylbenzydrylamine PS resin (Rink amide MBHA PS Resin) and 4-[(((2,4-dimethoxyphenyl)Fmoc-aminomethyl)phenoxyacetamido)aminomethyl]-benzhydrylamine PS resin (Rink amide BHA PS resin). Preferably, the support is derived from polystyrene crosslinked with, most preferably 1 to 5% of, divinylbenzene and functionalized by means of the 4-(((2,4-dimethoxyphenyl)Fmoc-aminomethyl)phenoxy-acetamido) linker.

[0029] Type 2 linkers are designed to eventually release the carboxyl group under acidic conditions. Linkers of this kind form acid-labile esters with the carboxyl group of the amino acid, usually acid-labile benzyl, benzhydryl and trityl esters; examples of such linker structures include 3-methoxy-4-hydroxymethylphenoxy (Sasrin® linker), 4-(2,4-dimethoxyphenyl-hydroxymethyl)-phenoxy (Rink linker), 4-(4-hydroxymethyl-3-methoxyphenoxy)butyric acid (HMPB linker), trityl and 2-chlorotrityl. Preferably, the support is derived from polystyrene crosslinked with, most preferably 1 to 5% of, divinylbenzene and functionalized by means of the 2-chlorotrityl linker.

[0030] When carried out as parallel array syntheses, the processes of the invention can be advantageously carried out as described hereinbelow, but it will be immediately apparent to those skilled in the art, how these procedures will have to be modified in case it is desired to synthesize one single compound of the invention.

[0031] A number of reaction vessels (normally 24 to 192, typically 96) equal to the total number of compounds to be synthesized by the parallel method are loaded with 25 to 1000 mg, preferably 100 mg, of the appropriate functionalized solid support, which is preferably derived from polystyrene cross-linked with 1 to 3% of divinylbenzene, or from Tentagel® resin.

[0032] The solvent to be used must be capable of swelling the resin and includes, but is not limited to, dichloromethane (DCM), dimethylformamide (DMF), N-methylpyrrolidone (NMP), dioxane, toluene, tetrahydrofuran (THF), ethanol (EtOH), trifluoroethanol (TFE), isopropylalcohol and the like. Solvent mixtures containing as at least one component a polar solvent (e. g. 20% TFE/DCM, 35% THF/NMP) are beneficial for ensuring high reactivity and solvation of the resin-bound peptide chains (G. B. Fields et al., J. Am. Chem. Soc. 1991, 113, 4202-4207).

[0033] With the development of various linkers, that release the C-terminal carboxylic acid group under mild acidic conditions not affecting acid-labile groups protecting functional groups in the side chain(s), considerable progresses have been made in the synthesis of protected peptide fragments. The 2-methoxy-4-hydroxy-benzylalcohol-derived linker (Sasrin® linker, Mergler et al., Tetrahedron Lett. 1988, 29, 4005-4008) is cleavable with diluted trifluoroacetic acid (0.5 to 1% TFA in DCM) and is stable to Fmoc deprotection conditions during the peptide synthesis, Boc/tBu-based additional protecting groups being compatible with this protection scheme. Other linkers suitable for the processes of the invention include the super acid labile 4-(2,4-dimethoxyphenyl-hydroxymethyl)-phenoxy linker (Rink linker, Rink, H. Tetrahedron Lett. 1987, 28, 3787-3790), where the removal of the peptide requires 10% acetic acid in DCM or 0.2% trifluoroacetic acid in DCM; the 4-(4-hydroxymethyl-3-methoxyphenoxy)butyric acid-derived linker (HMPB-linker, Flörsheimer et al., Peptides 1991,1990, 131), which is also cleaved with 1% TFA/DCM, in order to yield a peptide fragment containing all acid labile side-chain protective groups; and the 2-chlorotritylchloride linker (Barlos et al., Tetrahedron Lett. 1989, 30, 3943-3946), which allows the peptide detachment using a mixture of glacial acetic acid/trifluoroethanol/DCM (1:2:7) for 30 min.

[0034] Suitable protecting groups for amino acids and, respectively, for their residues are, for example:

- For the amino group (as is present, e. g., also in the side-chain of lysine):

  Cbz
  benzyloxycarbonyl
  Boc
  tert.-butyloxycarbonyl
  Fmoc

9-fluorenylmethoxycarbonyl
Alloc
allyloxycarbonyl
Teoc
trimethylsilylethoxycarbonyl
Tcc
trichloroethoxycarbonyl
Nps
o-nitrophenylsulfonyl
Trt
triphenymethyl or trityl

- For the carboxyl group (as is present, e. g., also in the side-chains of aspartic acid and glutamic acid) by conversion into esters with the alcohol components:

  tBu
  tert.-butyl
  Bn
  benzyl
  Me
  methyl
  Ph
  phenyl
  Pac
  phenacyl
  allyl
  Tse
  trimethylsilylethyl
  Tce
  trichloroethyl

- For the guanidino group (as is present, e. g., in the side-chain of arginine):

  Pmc
  2,2,5,7,8-pentamethylchroman-6-sulfonyl
  Ts
  tosyl (i. e. p-toluenesulfonyl)
  Cbz
  benzyloxycarbonyl
  Pbf
  pentamethyldihydrobenzofuran-5-sulfonyl

- For the hydroxy group (as is present, e. g., in the side-chains of threonine and serine):

  tBu
  tert.-butyl
  Bn

benzyl
Trt
trityl

- For the mercapto group (as is present, e. g., in the side-chain of cysteine):

  Acm
  acetamidomethyl
  tBu
  tert.-butyl
  Bn
  benzyl
  Trt
  trityl
  Mtr
  4-methoxytrityl

[0035] The Fmoc-protected amino acid derivatives are preferably used as the building blocks for the construction of the template-fixed beta-hairpin loop mimetics of the invention. For the deprotection, i. e. the cleaving off of the Fmoc group, 20% piperidine in DMF or 2% DBU/2% piperidine in DMF can be used.

[0036] The quantity of the reactant, i. e. of the amino acid derivative, is usually from 1 to 20 equivalents based on the milliequivalents per gram (meq/g) loading of the functionalized solid support (typically 0.1 to 2.85 meq/g for polystyrene resins) originally weighed into the reaction tube. Additional equivalents of reactant can be used, if required, to drive the reaction to completion in a reasonable time. The reaction tubes, in combination with the holder block and the manifold, are reinserted into the reservoir block, and the apparatus is fastened together. Gas flow through the manifold is initiated to provide a controlled environment, for example, nitrogen, argon, air and the like. The gas flow may also be heated or chilled prior to flowing through the manifold. Heating or cooling of the reaction wells is achieved by heating the reaction block or cooling externally with isopropanol/dry ice and the like to bring about the desired synthetic reactions. Agitation is achieved by shaking or magnetic stirring (within the reaction tube). The preferred workstations (without, however, being limited thereto) are Labsource's Combi-chem station and MultiSyn Tech's Syro synthesizer.

[0037] Amide bond formation requires the activation of the alpha-carboxyl group for the acylation step. When this activation is being carried out by means of the commonly used carbodiimides, such as dicyclohexylcarbodiimide (DCC, Sheehan et al., J. Am. Chem. Soc. 1955, 77, 1067-1068) or diisopropylcarbo (DIC, Sarantakis et al., Biochem. Biophys. Res. Commun. 1976, 73, 336-342), the resulting dicyclohexylurea and diisopropylurea is insoluble and, respectively, soluble in the solvents generally used. In a variation of the carbodiimide method, 1-hydroxybenzotriazole (HOBt, König et al., Chem. Ber 1970, 103, 788-798) is included as an additive to the coupling mixture. HOBt prevents dehydration, suppresses racemization of the activated amino acids and acts as a catalyst to improve the sluggish coupling reactions. Certain phosphonium reagents have been used as direct

coupling reagents, such as benzotriazol-1-yl-oxy-tris-(dimethylamino)-phosphonium hexafluorophosphate (BOP, Castro et al., Tetrahedron Lett. 1975, 14, 1219-1222; Synthesis, 1976, 751-752), benzotriazol-1-yl-oxy-tris-pyrrolidino-phosphonium hexaflurophoshate (Py-BOP, Coste et al., Tetrahedron Lett. 1990, 31, 205-208) or 2-(1H-benzotriazol-1-yl)-1,1,3,3-tetramethyluronium tetrafluoroborate (TBTU) or hexafluorophosphate (HBTU, Knorr et al., Tetrahedron Lett. 1989, 30, 1927-1930); these phosphonium reagents are also suitable for in situ formation of HOBt esters with the protected amino acid derivatives. More recently, diphenoxyphosphoryl azide (DPPA), O-(7-azabenzotriazol-1-yl)-N,N,N',N'-tetramethyluronium tetrafluoroborate (TATU) or O-(7-aza-benzotriazol-1-yl)-N,N,N',N'-tetramethyluronium hexafluorophosphate (HATU) / 7-aza-1-hydroxy benzotriazole (HOAt, Carpino et al., Tetrahedron Lett. 1994, 35, 2279-2281) have also been used as coupling reagents.

[0038] Due to the fact, that near-quantitative coupling reactions are essential, it is desirable to have experimental evidence for completion of the reactions. The ninhydrin test (Kaiser et al., Anal. Biochemistry 1970, 34, 595), where a positive colorimetric response to an aliquot of resin-bound peptide indicates qualitatively the presence of the primary amine, can easily and quickly be performed after each coupling step. Fmoc chemistry allows the spectrophotometric detection of the Fmoc chromophore, when it is released with the base (Meienhofer et al., Int. J. Peptide Protein Res. 1979, 13, 35-42).

[0039] The resin-bound intermediate within each reaction tube is washed free of excess of retained reagents, of solvents, and of by-products by repetitive exposure to pure solvent(s) by one of the two following methods:

1) The reaction wells are filled with solvent (preferably 5 ml), the reaction tubes, in combination with the holder block and the manifold, are immersed, agitated for 5 to 300 minutes, preferably 15 minutes, and drained by gravity followed by gas pressure applied through the manifold inlet (while closing the outlet) to expel the solvent.

2) The manifold is removed from the holder block, aliquots of solvent (preferably 5 ml) are dispensed through the top of the reaction tubes and drained by gravity through a filter into a receiving vessel, such as a test tube or vial.

[0040] Both of the above washing procedures are repeated up to about 50 times (preferably about 10 times), monitoring the efficiency of reagent, solvent, and byproduct removal by methods such as TLC, GC, or inspection of the washings.

[0041] The above-described procedure of reacting the resin-bound compound with reagents within the reaction wells, followed by removal of excess reagents, by-products, and solvents, is repeated with each successive transformation, until the final resin-bound fully protected linear peptide has been obtained.

[0042] Before this fully protected linear peptide is detached from the solid support, it is possible, if desired, to selectively deprotect one or several protected functional group(s)

present in the molecule and to appropriately substitute the reactive group(s) thus liberated. To this effect, the functional group(s) in question must initially be protected by a protecting group, which can be selectively removed without affecting the remaining protecting groups present. Alloc (allyloxycarbonyl) is an example for such an amino protecting group, which can be selectively removed, e. g. by means of Pd^0 and phenylsilane in CH_2Cl_2, without affecting the remaining protecting groups, such as Fmoc, present in the molecule. The reactive group thus liberated can then be treated with an agent suitable for introducing the desired substituent. Thus, for example, an amino group can be acylated by means of an acylating agent corresponding to the acyl substituent to be introduced.

[0043] Before this fully protected linear peptide is detached from the solid support, it is also possible, if desired, to form an interstrand linkage between side-chains of appropriate amino acid residues at positions 2 and 10.

[0044] Interstrand linkages, as discussed above, can be disulfide bridges, formed between cysteine residues at opposite positions of the beta-strand, or lactam bridges, formed between a glutamic acid residue and a lysine residue located at opposite beta-strand positions. The formation of such interstrand linkages can be effected by methods well known in the art.

[0045] For the formation of disulfide bridges, preferably a solution of 10 eq of iodine in DMF or a mixture of CH_2Cl_2 / MeOH is applied for 1.5 h and then for another 3 h with a fresh iodine solution after filtering of the iodine solution, or a mixture of DMSO and acetic acid solution, buffered with 5% $NaHCO_3$ to pH 5-6, for 4 h, or water, adjusted to pH 8 with ammonium hydroxide solution, with stirring for 24 h, or ammonium acetate buffer, adjusted to pH 8, in the presence of air, or a solution of NMP and tri-n-butylphosphine (preferably 50 eq).

[0046] The detachment of the fully protected linear peptide from the solid support is achieved by immersion of the reaction tubes, in combination with the holder block and the manifold, in reaction wells containing a solution of the cleavage reagent (preferably 3 to 5 ml). Gas flow, temperature control, agitation and reaction monitoring are implemented as described above and as desired to effect the detachment reaction. The reaction tubes, in combination with the holder block and the manifold, are disassembled from the reservoir block and raised above the solution level, but below the upper lip of the reaction wells, and gas pressure is applied through the manifold inlet (while closing the outlet) to efficiently expel the final product solution into the reservoir wells. The resin remaining in the reaction tubes is then washed 2 to 5 times as above with 3 to 5 ml of an appropriate solvent to extract (wash out) as much of the detached product as possible. The product solutions thus obtained are combined, taking care to avoid cross-mixing. The individual solutions/extracts are then manipulated as needed to isolate the final compounds. Typical manipulations include, but are not limited to, evaporation, concentration, liquid/liquid extraction, acidification, basification, neutralization or additional reactions in solution.

[0047] The solutions containing the fully protected linear peptide derivatives, which have been cleaved off from the solid support and neutralized with a base, are evaporated. The cyclization is then affected in solution, using solvents, such as DCM, DMF, dioxane, THF and the like. Various coupling reagents, which were mentioned earlier, can be used for the

cyclization. The duration of the cyclization is about 6-48 hours, preferably about 16 hours. The progress of the reaction is followed, e. g. by RP-HPLC (Reverse Phase High Performance Liquid Chromatography). Then the solvent is removed by evaporation, the fully protected cyclic peptide derivative is dissolved in a solvent not miscible with water, such as DCM, and the solution is extracted with water or a mixture of water-miscible solvents, in order to remove any excess of the coupling reagent.

[0048] Finally, the fully protected peptide derivative is treated with 95% of TFA, 2.5% of H_2O and 2.5% of TIS or another combination of scavengers for affecting the cleavage of the protecting groups. The cleavage reaction time is commonly 30 minutes to 12 hours, preferably about 2.5 hours. The volatiles are evaporated to dryness, and the crude peptide is dissolved in 20% AcOH in water, which is extracted with isopropyl ether or other solvents suitable therefor. The aqueous layer is collected and evaporated to dryness, and the fully deprotected cyclic peptide derivative of the formula I is obtained as end-product.

[0049] Alternatively, the detachment from the solid support, cyclization and complete deprotection of the fully protected peptide can be achieved manually in glass vessels.

[0050] Depending on its purity, the peptide derivative can be used directly for biological assays, or it has to be further purified, for example by preparative HPLC.

[0051] It is possible, if desired, to convert a fully deprotected product of the formula I in free form thus obtained into a pharmaceutically acceptable salt or to convert a pharmaceutically acceptable, or unacceptable, salt thus obtained into the corresponding compound of the formula I in free form or into a different, pharmaceutically acceptable, salt. Any of these operations can be carried out by methods well known in the art.

[0052] The starting materials of the formula II shown in claims 40 and 41, the pre-starting materials therefor, and the preparation of these starting and pre-starting materials are described in WO-A-2002070547.

[0053] The beta-hairpin peptidomimetics of the invention can be used in a wide range of applications, where cancer, inflammatory diseases, pulmonary diseases, infections, immunological diseases, cardiovascular diseases or neurodegenerative diseases are mediated by, or resulting from, serine protease activity. For the control or prevention of a given illness or disease amenable to treatment with protease inhibitors, the beta-hairpin peptidomimetics may be administered per se or applied as an appropriate formulation together with carriers, diluents or excipients well known in the art.

[0054] When used to treat or prevent diseases, such as pulmonary emphysema, rheumatoid arthritis, osteoarthritis, atherosclerosis, psoriaris, cystic fibrosis, multiple sclerosis, adult respiratory distress syndrome, pancreatitis, asthma, allergic rhinitis, inflammatory dermatoses, postangioplasty restenosis, cardiac hypertrophy, heart failure or cancer, such as, but not limited to, breast cancer or cancer related to angiogenesis or metastasis, the beta-hairpin peptidomimetics can be administered singly, as mixtures of several beta-hairpin peptidomimetics, in combination with other antiinflammatory, antimicrobial or anticancer agents and/or in combination with other pharmaceutically active

agents. The beta-hairpin peptidomimetics can be administered per se or as pharmaceutical compositions.

[0055] Pharmaceutical compositions comprising beta-hairpin peptidomimetics of the invention may be manufactured by means of conventional mixing, dissolving, granulating, coated tablet-making, levigating, emulsifying, encapsulating, entrapping or lyophilizing processes. Pharmaceutical compositions may be formulated in conventional manner using one or more physiologically acceptable carriers, diluents, excipients or auxiliaries, which facilitate processing of the active beta-hairpin peptidomimetics into preparations, which can be used pharmaceutically. The proper formulation depends on the method of administration chosen.

[0056] For topical administration, the beta-hairpin peptidomimetics of the invention may be formulated as solutions, gels, ointments, creams, suspensions, etc., as are well-known in the art.

[0057] Systemic formulations include those designed for administration by injection, e. g. subcutaneous, intravenous, intramuscular, intrathecal or intraperitoneal injection, as well as those designed for transdermal, transmucosal, oral or pulmonary administration.

[0058] For injections, the beta-hairpin peptidomimetics of the invention may be formulated in adequate solutions, preferably in physiologically compatible buffers, such as Hink's solution, Ringer's solution, or physiological saline buffer. The solutions may contain formulatory agents, such as suspending, stabilizing and/or dispersing agents. Alternatively, the beta-hairpin peptidomimetics of the invention may be in powder form for combination with a suitable vehicle, e. g. sterile pyrogen-free water, before use.

[0059] For transmucosal administration, penetrants appropriate to the barrier to be permeated are used in the formulation as known in the art.

[0060] For oral administration, the beta-hairpin peptidomimetics of the invention can be readily formulated by combining them with pharmaceutically acceptable carriers well known in the art. Such carriers enable the beta-hairpin peptidomimetics of the invention to be formulated as tablets, pills, dragees, capsules, liquids, gels, syrups, slurries, suspensions, etc. for oral ingestion by a patient to be treated. For oral formulations, such as powders, capsules and tablets, suitable excipients include fillers, such as sugars, e. g. lactose, sucrose, mannitol and sorbitol; cellulose preparations, such as maize starch, wheat starch, rice starch, potato starch, gelatin, gum tragacanth, methyl cellulose, hydroxypropylmethyl cellulose, sodium carboxymethyl cellulose or polyvinylpyrrolidone (PVP); granulating agents; and binding agents. If desired, desintegrating agents may be added, such as cross-linked polyvinylpyrrolidones, agar, or alginic acid or a salt thereof, such as sodium alginate. If desired, solid dosage forms may be sugar-coated or enteric-coated using standard techniques.

[0061] For oral liquid preparations, such as suspensions, elixirs and solutions, suitable carriers, excipients or diluents include water, glycols, oils, alcohols, etc.. In addition, flavoring agents, preservatives, coloring agents and the like may be added.

[0062] For buccal administration, the compositions may take the form of tablets, lozenges, etc., formulated as usual.

[0063] For administration by inhalation, the beta-hairpin peptidomimetics of the invention are conveniently delivered in the form of an aeorosol spray from pressurized packs or a nebulizer, with the use of a suitable propellant, e. g. dichlorodifluoromethane, trichlorofluoromethane, carbon dioxide or another suitable gas. In the case of a pressurized aerosol, the dose unit may be determined by providing a valve to deliver a metered amount. Capsules and cartridges of, e. g., gelatin for use in an inhaler or insufflator may be formulated containing a powder mix of the beta-hairpin peptidomimetic of the invention and a suitable powder base, such as lactose or starch.

[0064] The compounds may also be formulated in rectal or vaginal compositions, such as suppositories, together with appropriate suppository bases, such as cocoa butter or other glycerides.

[0065] In addition to the formulations described previously, the beta-hairpin peptidomimetics of the invention may also be formulated as depot preparations. Such long acting formulations may be administered by implantation (e. g. subcutaneously or intramuscularly) or by intramuscular injection. For the manufacture of such depot preparations, the beta-hairpin peptidomimetics of the invention may be formulated with suitable polymeric or hydrophobic materials (e. g. as an emulsion in an acceptable oil) or ion exchange resins, or as sparingly soluble salts.

[0066] In addition, other pharmaceutical delivery systems may be employed, such as liposomes and emulsions well known in the art. Certain organic solvents, such as dimethylsulfoxide, may also be employed. Additionally, the beta-hairpin peptidomimetics of the invention may be delivered using a sustained-release system, such as semipermeable matrices of solid polymers containing the therapeutic agent. Various sustained-release materials have been established and are well known in the art. Sustained-release capsules may, depending on their chemical nature, release the compound for a few weeks or up to over 100 days. Depending on the chemical nature and the biological stability of the therapeutic agent, additional strategies for protein stabilization may be employed.

[0067] As the beta-hairpin pepdidomimetics of the invention may contain charged residues, they may be included in any of the above-described formulations as such or as pharmaceutically acceptable salts. Pharmaceutically acceptable salts tend to be more soluble in aqueous and other protic solvents than the corresponding free forms.

[0068] The beta-hairpin peptidomimetics of the invention, or compositions thereof, will generally be used in an amount effective to achieve the intended purpose. It is to be understood, that the amount used will depend on a particular application.

[0069] For topical administration, to treat or prevent diseases amenable to beta-hairpin mimetics, a therapeutically effective dose can be determined using, for example, the in vitro assays provided in the examples. The treatment may be applied, while the disease is visible or even, when it is not visible. An ordinarily skilled expert will be able to determine

therapeutically effective amounts to treat topically diseases without undue experimentation.

[0070] For systemic administration, a therapeutically effective dose can be estimated initially from in vitro assays. For example, a dose can be formulated in animal models to achieve a circulating beta-hairpin peptidomimetic concentration range, that includes the IC_{50} as determined in the cell culture. Such information can be used to more accurately determine useful doses in humans.

[0071] Initial dosages can also be determined from in vivo data, e. g. animal models, using techniques well known in the art. One having ordinary skill in the art could readily optimize the administration to humans based on animal data.

[0072] Dosage amounts for application as serine protease inhibitory agents may be adjusted individually to provide plasma levels of the beta-hairpin peptidomimetics of the invention, which are sufficient to maintain the therapeutic effect. Therapeutically effective serum levels may be achieved by administering multiple doses each day.

[0073] In cases of local administration or selective uptake, the effective local concentration of the beta-hairpin peptidomimetics of the invention may not be related to the plasma concentration. One having ordinary skill in the art will be able to optimize therapeutically effective local dosages without undue experimentation.

[0074] The amount of beta-hairpin peptidomimetics administered will, of course, depend on the subject being treated, on the subject's weight, the severity of the affliction, the manner of administration and the judgement of the prescribing physician.

[0075] Normally, a therapeutically effective dose of the beta-hairpin peptidomimetics described herein will provide a therapeutic benefit without causing substantial toxicity.

[0076] The toxicity of the beta-hairpin peptidomimetics of the invention can be determined by standard pharmaceutical procedures in cell cultures or experimental animals, e. g. by determining the LD_{50} (the dose lethal to 50% of the population) or the LD_{100} (the dose lethal to 100% of the population). The dose ratio between toxic and therapeutic effect is the therapeutic index. Compounds, which exhibit high therapeutic indices, are preferred. The data obtained from these cell culture assays and animal studies can be used in formulating a dosage range, that is not toxic for use in humans. The dosage of the beta-hairpin peptidomimetics of the invention lies preferably within a range of circulating concentrations, that include the effective dose with little or no toxicity. The dosage may vary within the range, depending on the dosage form employed and the route of administration utilized. The exact formulation, route of administration and dose can be chosen by the individual physician in view of the patient's condition (see, e. g., Fingl et al., 1975, The Pharmacological Basis of Therapeutics, Ch. 1, p. 1).

[0077] The following examples illustrate the invention in more detail, but are not intended to limit its scope in any way.

Examples

[0078] The following abbreviations are used in the examples:

HBTU
: 1-benzotriazol-1-yl-tetramethyluronium hexafluorophosphate (Knorr et al., Tetrahedron Lett. 1989, 30, 1927-1930)

HOBt
: 1-hydroxybenzotriazole

DIEA
: diisopropylethylamine

HATU
: O-(7-aza-benzotriazole-1-yl)-N,N,N',N'-tetramethyluronium hexafluorophosphate (Carpino et al., Tetrahedron Lett. 1994, 35, 2279-2281)

1. Peptide synthesis

Coupling of the first protected amino acid residue to the resin

[0079] 0.5 g of 2-chlorotritylchloride resin (Barlos et al., Tetrahedron Lett. 1989, 30, 3943-3946) (0.83 mmol/g, 0.415 mmol) were filled into a dried flask. The resin was suspended in CH_2Cl_2 (2.5 ml) and allowed to swell at room temperature under constant stirring for 30 min. The resin was treated with 0.415 mmol (1 eq) of the first suitably protected amino acid (see below) and 284 microliters (4 eq) of DIEA in CH_2Cl_2 (2.5 ml), and the mixture was shaken at 25°C for 4 h. The resin colour changed to purple, while the solution remained yellowish. The resin was shaken (CH_2Cl_2 / MeOH / DIEA, 17 : 2 : 1; 30 ml) for 30 min, then washed with CH_2Cl_2 (1x), DMF (1x), CH_2Cl_2 (1x), MeOH (1x), CH_2Cl_2 (1x), MeOH (1x), CH_2Cl_2 (2x) and Et_2O (2x) and dried under vacuum for 6 h. The loading was typically 0.6 to 0.7 mmol/g.

[0080] The preloaded resins Fmoc-Pro-2-chlorotritylresin, Fmoc-Asp(OtBu)-2-chlorotritylresin, Fmoc-Leu-2-chlorotritylresin, Fmoc-Glu(OtBu)-2-chlorotritylresin, Fmoc-Ser(OtBu)-2-chlorotritylresin and Fmoc-Ile-2-chlorotritylresin were prepared.

Synthesis of the fully protected peptide fragment

[0081] The synthesis was carried out with a Syro peptide synthesizer (MultiSyn Tech) using 24 to 96 reaction vessels. In each vessel were placed 60 mg (weight of the resin before loading) of the above resin. The following reaction cycles were programmed and carried out:

Step	Reagent	Time
1	CH_2Cl_2, wash and swell (manual)	3 × 1 min
2	DMF, wash and swell	1 × 5 min
3	40% piperidine / DMF	1 × 5 min
4	DMF, wash	5 × 2 min
5	5 eq Fmoc amino acid / DMF	
	+ 5 eq HBTU	
	+ 5 eq HOBt	
	+ 5 eq DIEA	1 × 120 min
6	DMF, wash	4 × 2 min
7	CH_2Cl_2, wash (at the end of the synthesis)	3 × 2 min

[0082] The steps 3 to 6 were repeated to add each amino acid.

[0083] After the synthesis of the fully protected peptide fragment, either Procedure A or Procedure B was adopted, depending on whether or not a disulfide interstrand linkage was to be formed.

Procedure A: Cyclization, followed by work-up of backbone-cyclized peptide

Cleavage of the fully protected peptide fragment

[0084] After the completion of the synthesis, the resin was suspended in 1 ml (0.39 mmol) of 1% TFA in CH_2Cl_2 (vlv) for 3 min and filtered off. The filtrate was neutralized with 1 ml (1.17 mmol, 3 eq) of 20% DIEA in CH_2Cl_2 (v/v). This procedure was repeated twice to ensure the completion of the cleavage.

[0085] An aliquot (200 microliters) of the filtrate was fully deprotected with 0.5 ml of a mixture containing 95% of TFA, 2.5% of water and 2.5% of triisopropylsilane (TIS) and analysed by reverse phase LC / MS to monitor the efficiency of the linear peptide synthesis.

Cyclization of the linear peptide

[0086] The fully protected linear peptide was dissolved in DMF (8 ml; concentration 10 mg/ml). 2 eq of HATU (0.72 mmol) in 1 ml of DMF and 4 eq of DIEA (1.44 mmol) in 1 ml of

DMF were added, and the mixture was stirred at room temperature for 16 h. The volatiles were evaporated to dryness. The crude cyclised peptide was dissolved in 7 ml of CH_2Cl_2 and extracted with 10% acetonitrile in water (4.5 ml) three times. The CH_2Cl_2 layer was evaporated to dryness.

Deprotection and purification of the cyclic peptide

[0087] The cyclic peptide obtained was dissolved in 3 ml of a mixture containing 95% of TFA, 2.5% of water and 2.5% of triisopropylsilane (TIS). The mixture was left to stand at 20°C for 2.5 h and then concentrated under vacuum. The crude peptide was dissolved in 20% AcOH in water (7 ml) and extracted with diisopropylether (4 ml) three times. The aqueous layer was collected and evaporated to dryness, and the residue was purified by preparative reverse phase LC / MS. After lyophilisation, the product was obtained as a white powder and analysed by LC / MS. The analytical data comprising purity after preparative H PLC and ESI-MS are shown in table 1. The analytical HPLC retention times (RT; min) were determined using a Jupiter Proteo 90A, 150 x 2.0 mm (cod. 00F4396-B0 - Phenomenex), with the solvents A (H_2O + 0.1 % TFA) and B (CH_3CN + 0.1% TFA) and the gradient 0 min: 95% A, 5% B; 20 min: 40% A, 60% B; 21-23 min: 0% A, 100% B; 23.1-30 min: 95% A, 5% B.

Procedure B: Cyclization, followed by work-up of backbone-cyclized peptide having a disulfide interstrand linkage

Formation of disulfide interstrand linkage

[0088] After the completion of the synthesis, the resin was swelled in 3 ml of dry DMF for 1 h. Then a solution of 10 eq of iodine in DMF (6 ml) was added to the reactor, followed by stirring for 1.5 h. The resin was filtered, and a fresh solution of iodine (10 eq) in DMF (6 ml) was added, followed by stirring for another 3 h. The resin was filtered and washed with DMF (3x) and CH_2Cl_2 (3x).

Backbone cyclization, cleavage and purification of the peptide

[0089] After the formation of the disulfide interstrand linkage, the resin was suspended in 1 ml (0.39 mmol) of 1% TFA in CH_2Cl_2 (*v/v*) for 3 min and filtered off. The filtrate was neutralized with 1 ml (1.17 mmol, 3 eq) of 20% DIEA in CH_2Cl_2 (*v/v*). This procedure was repeated twice to ensure the completion of the cleavage. The resin was washed with 2 ml of CH_2Cl_2. The CH_2Cl_2 layer was evaporated to dryness.The fully protected linear peptide was solubilised in 8 ml of dry DMF. Then 2 eq of HATU in dry DMF (1ml) and 4 eq of DIEA in dry DMF (1 ml) were added, followed by stirring for 16 h. The volatiles were evaporated

to dryness. The crude cyclised peptide was dissolved in 7 ml of CH_2Cl_2 and extracted with 10% acetonitrile in water (4.5 ml) three times. The CH_2Cl_2 layer was evaporated to dryness. To deprotect the peptide fully, 3 ml of TFA / TIS / H_2O (95 : 2.5 : 2.5) were added, and the mixture was kept for 2.5 h. The volatiles were evaporated to dryness, and the crude peptide was dissolved in 20% AcOH in water (7 ml) and extracted with diisopropylether (4 ml) three times. The aqueous layer was collected and evaporated to dryness, and the residue was purified by preparative reverse phase LC / MS. After lyophilisation, the product was obtained as a white powder and analysed by ESI-MS as described above. The analytical data comprising purity after preparative HPLC and ESI-MS are shown in table 1.

Examples 1 to 6, 9 to 45, 52 to 63, 65 to 67, 70, 71, 76 to 114, 129, 131 to 162, 179 to 182, 184 to 189 and 191 to 195

[0090] The peptides of these examples, which are listed in table 1, were synthesized starting with the amino acid Pro, which was grafted to the resin. The starting resin was Fmoc-Pro-2-chlorotritylresin prepared as described above. The linear peptides were synthesized on the solid support according to the procedure described above in the following sequence: Resin-Pro-^{D}Pro-P11-P10-P9-P8-P7-P6-P5-P4-P3-P2-P1.

[0091] In the examples 1 to 6, 9 to 45, 52 to 63, 65 to 67, 70, 71, 76 to 81, 83 to 103, 112 to 114, 129, 131, 133, 136 to 138, 140, 141, 143 to 146, 148, 150 to 153, 155, 157, 158, 160, 162, 179 to 182, 184 to 189 and 191 to 195, the peptide was cleaved from the resin, subjected to the disulfide bridge formation, cyclized, deprotected and purified as indicated in Procedure B.

[0092] In the examples 82, 149, 159 and 161, the peptide was cleaved from the resin as indicated in Procedure B. Then the crude product was solubilised in ammonium acetate buffer (0.1M; pH adjusted to 8; concentration 1 mg of crude product per ml). The mixture was stirred at room temperature in the presence of air. The reaction was monitored by reverse phase LC / MS. After the completion of the reaction, the mixture was evaporated to dryness, and the residue was purified by preparative reverse phase LC / MS. The cyclisation of the backbone was performed as indicated in Procedure A. To deprotect the peptide fully, 5 ml of TFA / H_2O / phenol / thioanisol / ethanedithiol (82.5 : 5 : 5 : 5 : 2.5) were added, and the mixture was kept for 5 h at room temperature. The peptide was precipitated by the addition of cold diethylether (10ml). After centrifugation, the supernatant phase was removed. The precipitate was washed three times with 5 ml of diethylether and purified by preparative reverse phase LC / MS. After lyophilisation, the product was obtained as a white powder and analysed by ESI-MS as described above.

[0093] In the examples 104 to 111, 132, 134, 135, 139, 142, 147, 154 and 156, the peptide was cleaved from the resin, cyclized, deprotected and purified as indicated in Procedure A.

[0094] The following HPLC retention times (min) were determined using the analytical method described above (Ex. = example):

Ex. 1 (15.37), Ex. 2 (11.54), Ex. 3 (7.82), Ex. 4 (8.62), Ex. 5 (16.51), Ex. 6 (13.67), Ex. 9 (5.82), Ex. 10 (7.98), Ex. 11 (8.38), Ex. 12 (6.80), Ex. 13 (7.41), Ex. 14 (6.20), Ex. 15 (8.68), Ex. 16 (9.82), Ex. 17 (5.59), Ex. 20 (7.32), Ex. 21 (8.66), Ex. 22 (8.68), Ex. 23 (12.66), Ex. 24 (8.67), Ex. 25 (7.53), Ex. 26 (9.02), Ex. 27 (8.06), Ex. 28 (9.62), Ex. 29 (8.78), Ex. 30 (10.49), Ex. 31 (5.50), Ex. 32 (7.45), Ex. 33 (8.39), Ex. 34 (10.16), Ex. 35 (9.04), Ex. 36 (10.98), Ex. 37 (7.56), Ex. 38 (9.29), Ex. 39 (8.32), Ex. 40 (10.11), Ex. 41 (7.23), Ex. 42 (8.83), Ex. 43 (7.92), Ex. 44 (9.87), Ex. 45 (8.26), Ex. 52 (6.20), Ex. 53 (8.68), Ex 54 (9.82), Ex. 55 (5.59), Ex. 56 (6.06), Ex. 57 (6.47), Ex. 58 (7.32), Ex. 59 (8.68), Ex. 60 (10.66), Ex. 61 (8.54), Ex. 62 (9.83), Ex. 63 (16.54), Ex. 65 (15.71), Ex. 66 (17.50), Ex. 67 (15.87), Ex. 70 (12.87), Ex. 71 (13.48), Ex. 76 (4.47), Ex. 77 (5.15), Ex. 78 (10.93), Ex. 79 (10.70), Ex. 80 (12.09), Ex. 81 (11.63), Ex. 82 (5.71), Ex. 83 (5.45), Ex. 84 (11.14), Ex. 85 (10.90), Ex. 86 (13.78), Ex. 87 (13.98), Ex. 88 (14.35), Ex. 89 (15.21), Ex. 90 (14.72), Ex. 91 (11.97), Ex. 92 (11.77), Ex. 93 (15.25), Ex. 94 (14.61), Ex. 95 (20.46), Ex. 96 (15.08), Ex. 97 (20.78), Ex. 98 (18.28), Ex. 99 (14.62), Ex. 100 (13.90), Ex. 101 (13.76), Ex. 102 (20.53), Ex. 103 (14.14), Ex. 104 (11.60), Ex. 105 (11.90), Ex. 106 (11.63), Ex. 107 (11.78), Ex. 108 (13.03), Ex. 109 (15.22), Ex. 110 (12.40), Ex. 111 (12.10), Ex. 112 (5.49), Ex. 113 (5.67), Ex. 114 (5.55), Ex. 129 (17.22), Ex. 131 (11.97), Ex. 132 (13.56), Ex. 133 (14.57), Ex. 134 (14.72), Ex. 135 (17.53), Ex. 136 (18.28), Ex. 137 (14.72), Ex. 138 (14.35), Ex. 139 (15.40), Ex. 140 (11.14), Ex. 141 (5.71), Ex. 142 (13.97), Ex. 143 (13.94), Ex. 144 (15.08), Ex. 145 (20.87), Ex. 146 (17.91), Ex. 147 (17.11), Ex. 148 (7.83), Ex. 149 (16.22), Ex. 150 (20.09), Ex. 151 (20.72), Ex. 152 (21.38), Ex. 153 (17.97), Ex. 154 (16.58), Ex. 155 (19.46), Ex. 156 (15.66), Ex. 157 (22.04), Ex. 158 (15.65), Ex. 159 (17.89), Ex. 160 (18.72), Ex. 161 (19.91), Ex. 162 (17.79), Ex. 179 (4.25), Ex. 180 (11.43), Ex. 181 (12.30), Ex. 182 (12.83), Ex. 184 (12.12), Ex. 185 (10.14), Ex. 186 (10.09), Ex. 187 (10.14), Ex. 188 (10.65), Ex. 189 (10.73), Ex. 191 (10.17), Ex. 192 (10.19), Ex. 193 (11.02), Ex. 194 (9.92), Ex. 195 (10.74).

Table 1

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
1	SEQ ID NO: 1	Phe	Cys	Thr	Lys	Ser	Glu	Pro	Pro	Ile	Cys	Thr	^{D}Pro^{L}Pro	95%	1385.7
2	SEQ ID NO: 2	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asp	^{D}Pro^{L}Pro	93%	1399.5
3	SEQ ID NO: 3	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	95%	1398.5
4	SEQ ID NO: 4	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1371.1
5	SEQ ID NO: 5	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1447.5
6	SEQ ID NO: 6	Tyr	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Thr	^{D}Pro^{L}Pro	95%	1401.7
9	SEQ ID NO: 9	4AmPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	92%	1386.9
10	SEQ ID NO: 10	Nle	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	93%	1337.8
11	SEQ ID NO: 11	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1363.8

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
12	SEQ ID NO: 12	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	95%	1432.7
13	SEQ ID NO: 13	2Cl-Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	95%	1474.5
14	SEQ ID NO: 14	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Ala	Ile	Cys	Arg	^{D}Pro^{L}Pro	93%	1380.5
15	SEQ ID NO: 15	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Nle	Cys	Ser	^{D}Pro^{L}Pro	95%	1371.8
16	SEQ ID NO: 16	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Ser	^{D}Pro^{L}Pro	95%	1411.6
17	SEQ ID NO: 17	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Gln	Cys	Arg	^{D}Pro^{L}Pro	95%	1421.6
18	SEQ ID NO: 18	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Tyr	Cys	Arg	^{D}Pro^{L}Pro	89%	1456.6
19	SEQ ID NO: 19	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Nle	Cys	Arg	^{D}Pro^{L}Pro	95%	1476.6
20	SEQ ID NO: 20	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Arg	^{D}Pro^{L}Pro	95%	1446.5

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
21	SEQ ID NO: 21	Phe	Cys	Thr	Lys	Ser	Glu	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1385.8
22	SEQ ID NO: 22	Ile	Cys	Thr	Nle	Ser	Asp	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	95%	1391.6
23	SEQ ID NO: 23	Phe	Cys	Thr	Nle	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1432.7
24	SEQ ID NO: 24	Phe	Cys	Thr	Lys	AlloThr	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1385.7
25	SEQ ID NO: 25	Phe	Cys	Thr	Lys	Dpr	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1370.9
26	SEQ ID NO: 26	Tyr	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1463.8
27	SEQ ID NO: 27	hPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	95%	1412.6
28	SEQ ID NO: 28	hPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Thr	^{D}Pro^{L}Pro	95%	1399.7
29	SEQ ID NO: 29	hPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asp	^{D}Pro^{L}Pro	95%	1413.6

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
30	SEQ ID NO: 30	hPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1461.7
31	SEQ ID NO: 31	4AmPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	91%	1413.8
32	SEQ ID NO: 32	4AmPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	93%	1462.7
33	SEQ ID NO: 33	Cha	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	94%	1404.8
34	SEQ ID NO: 34	Cha	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Thr	^{D}Pro^{L}Pro	95%	1391.7
35	SEQ ID NO: 35	Cha	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asp	^{D}Pro^{L}Pro	95%	1405.8
36	SEQ ID NO: 36	Cha	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1453.8
37	SEQ ID NO: 37	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	95%	1390.7
38	SEQ ID NO: 38	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Thr	^{D}Pro^{L}Pro	95%	1377.6

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
39	SEQ ID NO: 39	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asp	^{D}Pro^{L}Pro	95%	1391.6
40	SEQ ID NO: 40	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1439.6
41	SEQ ID NO: 41	Nle	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	95%	1364.7
42	SEQ ID NO: 42	Nle	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Thr	^{D}Pro^{L}Pro	93%	1351.7
43	SEQ ID NO: 43	Nle	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asp	^{D}Pro^{L}Pro	95%	1365.7
44	SEQ ID NO: 44	Nle	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1413.6
45	SEQ ID NO: 45	2Cl-Plie	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Asn	^{D}Pro^{L}Pro	95%	1432.6
52	SEQ ID NO: 52	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Ala	Ile	Cys	Arg	^{D}Pro^{L}Pro	93%	1380.7
53	SEQ ID NO: 53	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Nle	Cys	Ser	^{D}Pro^{L}Pro	95%	1371.8

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
54	SEQ ID NO: 54	Phe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Ser	^{D}Pro^{L}Pro	95%	1411.6
55	SEQ ID NO: 55	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Gln	Cys	Arg	^{D}Pro^{L}Pro	95%	1421.6
56	SEQ ID NO: 56	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Tyr	Cys	Arg	^{D}Pro^{L}Pro	89%	1456.5
57	SEQ ID NO: 57	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Nle	Cys	Arg	^{D}Pro^{L}Pro	94%	1406.6
58	SEQ ID NO: 58	Ile	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Arg	^{D}Pro^{L}Pro	95%	1446.5
59	SEQ ID NO: 59	Ile	Cys	Thr	Nle	Ser	Asp	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	95%	1391.6
60	SEQ ID NO: 60	Phe	Cys	Thr	Nle	Ser	Asp	Pro	Pro	Ile	Cys	Tyr	^{D}Pro^{L}Pro	95%	1432.7
61	SEQ ID NO: 61	Trp	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1410.9
62	SEQ ID NO: 62	1-Nal	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Ile	Cys	Ser	^{D}Pro^{L}Pro	95%	1421.9

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
63	SEQ ID NO: 63	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Nle	Cys	Tyr	^{D}Pro^{L}Pro	95%	1439
65	SEQ ID NO: 65	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Tyr	Cys	Tyr	^{D}Pro^{L}Pro	95%	1489.5
66	SEQ ID NO: 66	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Tyr	^{D}Pro^{L}Pro	95%	1479.6
67	SEQ ID NO: 67	Chg	Cys	Thr	Lys	AlloThr	Asp	Pro	Pro	Tyr	Cys	Tyr	^{D}Pro^{L}Pro	95%	1503.6
70	SEQ ID NO: 70	4AmPhe	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Asn	^{D}Pro^{L}Pro	92%	1454.5
71	SEQ ID NO: 71	Chg	Cys	Thr	Lys	Ser	Asp	Pro	Pro	Cha	Cys	Arg	^{D}Pro^{L}Pro	95%	1472.6
76	SEQ ID NO: 76	Ile	Cys	Thr	Ala	Ser	Leu	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1369.3
77	SEQ ID NO: 77	Nle	Cys	Thr	Thr	Ser	Ile	Pro	Pro	Tyr	Cys	Tyr	^{D}Pro^{L}Pro	95%	1434.3
78	SEQ ID NO: 78	Nle	Cys	Thr	Abu	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1383.6

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
79	SEQ ID NO: 79	Nle	Cys	Thr	Ala	Ser	Nle	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1369.8
80	SEQ ID NO: 80	Nle	Cys	Thr	Ala	Ser	Aoc	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1397.6
81	SEQ ID NO: 81	Nle	Cys	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1425.6
82	SEQ ID NO: 82	Nle	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1409.5
83	SEQ ID NO: 83	Nle	Cys	Thr	Ala	Ser	hLeu	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1383.6
84	SEQ ID NO: 84	Nle	Cys	Thr	Ala	Ser	Chg	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1395.7
85	SEQ ID NO: 85	Nle	Cys	Thr	Ala	Ser	t-BuA	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1383.6
86	SEQ ID NO: 86	Nle	Cys	Ala	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1340.1
87	SEQ ID NO: 87	Nle	Cys	Abu	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1354.0

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Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
88	SEQ ID NO: 88	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro(4NHCOPhe)	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1488.6
89	SEQ ID NO: 89	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Phe	Cys	Tyr	^{D}Pro^{L}Pro	88%	1388.7
90	SEQ ID NO: 90	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	95%	1353.6
91	SEQ ID NO: 91	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1334.5
92	SEQ ID NO: 92	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Arg	^{D}Pro^{L}Pro	56%	1362.6
93	SEQ ID NO: 93	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Ser	^{D}Pro^{L}Pro	95%	1293.7
94	SEQ ID NO: 94	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Nle	^{D}Pro^{L}Pro	95%	1319.5
95	SEQ ID NO: 95	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	2-Nal	^{D}Pro^{L}Pro	94%	1404.0
96	SEQ ID NO: 96	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	2Cl-Phe	^{D}Pro^{L}Pro	94%	1387.8

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
97	SEQ ID NO: 97	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Cha	^{D}Pro^{L}Pro	95%	1359.8
98	SEQ ID NO: 98	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Phg	^{D}Pro^{L}Pro	95%	1359.9
99	SEQ ID NO: 99	Aoc	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	93%	1397.4
100	SEQ ID NO: 100	hLeu	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1383.4
101	SEQ ID NO: 101	Chg	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	87%	1395.6
102	SEQ ID NO: 102	OctG	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1425.5
103	SEQ ID NO: 103	hPhe	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1417.5
104	SEQ ID NO: 104	Nle	Glu	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Lys	Tyr	^{D}Pro^{L}Pro	95%	1422.8
105	SEQ ID NO: 105	Nle	Glu	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Arg	Tyr	^{D}Pro^{L}Pro	95%	1450.9

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
106	SEQ ID NO: 106	Nle	Thr	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Lys	Tyr	^{D}Pro^{L}Pro	95%	1394.7
107	SEQ ID NO: 107	Nle	Gln	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Arg	Tyr	^{D}Pro^{L}Pro	90%	1449.8
108	SEQ ID NO: 108	Nle	Thr	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Met	Tyr	^{D}Pro^{L}Pro	96%	1397.7
109	SEQ ID NO: 109	Nle	Gln	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Thr	Tyr	^{D}Pro^{L}Pro	95%	1394.7
110	SEQ ID NO: 110	Nle	Thr	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Gln	Tyr	^{D}Pro^{L}Pro	81%	1394.6
111	SEQ ID NO: 111	Nle	Gln	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Ser	Tyr	^{D}Pro^{L}Pro	95%	1380.7
112	SEQ ID NO: 112	Nle	Cys	Thr	Ala	Ser	C_5al	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	85%	1413.8
113	SEQ ID NO: 113	Nle	Cys	Thr	Ala	Ser	Leu	Pro	Pro	Tyr	Cys	Tyr	^{D}Pro^{L}Pro	95%	1404.7
114	SEQ ID NO: 114	Ile	Cys	Thr	Ala	Ser	Leu	Pro	Pro	Tyr	Cys	Tyr	^{D}Pro^{L}Pro	95%	1404.7

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
129	SEQ ID NO: 129	Nle	Cys	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	91%	1390.4
131	SEQ ID NO: 131	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1334.5
132	SEQ ID NO: 132	hPhe	Glu	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Lys	Tyr	^{D}Pro^{L}Pro	95%	1470.6
133	SEQ ID NO: 133	Nle	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1440.5
134	SEQ ID NO: 134	hPhe	Thr	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Gln	Tyr	^{D}Pro^{L}Pro	95%	1442.5
135	SEQ ID NO: 135	Nle	Thr	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Gln	Tyr	^{D}Pro^{L}Pro	88%	1450.7
136	SEQ ID NO: 136	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Phg	^{D}Pro^{L}Pro	95%	1339.9
137	SEQ ID NO: 137	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	95%	1353.6
138	SEQ ID NO: 138	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro(4NHCOPhe)	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1488.6

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
139	SEQ ID NO: 139	Nle	Thr	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Gln	Tyr	^{D}Pro^{L}Pro	95%	1434.8
140	SEQ ID NO: 140	Nle	Cys	Thr	Ala	Ser	Chg	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1395.7
141	SEQ ID NO: 141	Nle	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1409.5
142	SEQ ID NO: 142	hPhe	Gln	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Thr	Tyr	^{D}Pro^{L}Pro	91%	1406.5
143	SEQ ID NO: 143	hPhe	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	94%	1383.5
144	SEQ ID NO: 144	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	2Cl-Phe	^{D}Pro^{L}Pro	94%	1387.8
145	SEQ ID NO: 145	OctG	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	95%	1409.4
146	SEQ ID NO: 146	hPhe	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	95%	1401.5
147	SEQ ID NO: 147	OctG	Thr	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Gln	Tyr	^{D}Pro^{L}Pro	95%	1450.9

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Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
148	SEQ ID NO: 148	OctG	Cys	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1446.6
149	SEQ ID NO: 149	OctG	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1390.4
150	SEQ ID NO: 150	OctG	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1465.6
151	SEQ ID NO: 151	OctG	Cys	Thr	Ala	Ser	OctG	Pro	Pro(4NHCOPhe)	Gln	Cys	Gln	^{D}Pro^{L}Pro	94%	1565.7
152	SEQ ID NO: 152	hPhe	Cys	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	95%	1457.6
153	SEQ ID NO: 153	hPhe	Cys	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1438.5
154	SEQ ID NO: 154	OctG	Gln	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Thr	Tyr	^{D}Pro^{L}Pro	93%	1450.9
155	SEQ ID NO: 155	hPhe	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	90%	1441.5
156	SEQ ID NO: 156	OctG	Glu	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Lys	Tyr	^{D}Pro^{L}Pro	95%	1478.7

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
157	SEQ ID NO: 157	OctG	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Phe	^{D}Pro^{L}Pro	95%	1449.8
158	SEQ ID NO: 158	hPhe	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	94%	1422.7
159	SEQ ID NO: 159	OctG	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Gln	^{D}Pro^{L}Pro	93%	1430.0
160	SEQ ID NO: 160	OctG	Cys	Thr	Ala	Ser	Cha	Pro	Pro(4NHCOPhe)	Gln	Cys	Gln	^{D}Pro^{L}Pro	95%	1549.6
161	SEQ ID NO: 161	hPhe	Cys	Thr	Ala	Ser	OctG	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1473.4
162	SEQ ID NO: 162	hPhe	Cys	Thr	Ala	Ser	Cha	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	95%	1457.3
179	SEQ ID NO: 179	Nle	Cys	Thr	Ala	Ser	Ile	Pro	Pro	Gln	Cys	Tyr	^{D}Pro^{L}Pro	90%	1369.7
180	SEQ ID NO: 180	Ile	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	94%	1404.8
181	SEQ ID NO: 181	Ile	Cys	Thr	Lys	Ser	hPhe	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	92%	1452.6

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
182	SEQ ID NO: 182	Ile	Cys	Thr	Lys	Ser	Cha	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	95%	1444.6
184	SEQ ID NO: 184	Phe	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Ile	Cys	Arg	^{D}Pro^{L}Pro	95%	1438.6
185	SEQ ID NO: 185	Ile	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Arg	Cys	Arg	^{D}Pro^{L}Pro	95%	1447.5
186	SEQ ID NO: 186	Ile	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Lys	Cys	Arg	^{D}Pro^{L}Pro	95%	1419.9
187	SEQ ID NO: 187	Ile	Cys	Thr	Lys	Ser	Leu	Pro	Pro	His	Cys	Arg	^{D}Pro^{L}Pro	95%	1428.6
188	SEQ ID NO: 188	Ile	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Gln	Cys	Arg	^{D}Pro^{L}Pro	95%	1419.8
189	SEQ ID NO: 189	Ile	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Thr	Cys	Arg	^{D}Pro^{L}Pro	95%	1392.4
191	SEQ ID NO: 191	Leu	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Lys	Cys	Arg	^{D}Pro^{L}Pro	95%	1420.0
192	SEQ ID NO: 192	Nle	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Lys	Cys	Arg	^{D}Pro^{L}Pro	95%	1420.0

Example	Sequ. ID	P1	P2	P3	P4	P5	P6	P7	P8	P9	P10	P11	Template	Pur.[a)]	[M + H]
193	SEQ ID NO: 193	Cha	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Lys	Cys	Arg	^{D}Pro^{L}Pro	95%	1459.7
194	SEQ ID NO: 194	Tyr	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Lys	Cys	Arg	^{D}Pro^{L}Pro	95%	1469.6
195	SEQ ID NO: 195	Trp	Cys	Thr	Lys	Ser	Leu	Pro	Pro	Lys	Cys	Arg	^{D}Pro^{L}Pro	92%	1492.6

Examples 1 to 6, 9 to 45, 52 to 63, 65 to 67, 70, 71, 76 to 103, 112 to 114, 129, 131, 133, 136 to 138, 140, 141, 143 to 146, 148 to 153, 155, 157 to 162, 179 to 182, 184 to 189 and 191 to 195: Cys residues at P2 and P10 are linked by disulfide bridge.
a) Pur. = Purity of compound after preparative HPLC

2. Biological methods

2.1. Preparation of the peptide sample

[0095] The lyophilized peptide was weighed on a microbalance (Mettler MT5) and dissolved in sterile water to a final concentration of 1mM, unless stated otherwise. Stock solutions were kept at +4°C, light protected.

2.2. Enzymatic assays

[0096] The enzyme and substrate conditions are indicated in table 2. The kinetic measurements were made in a total reaction volume of 100 microliters in 96-well flat bottomed plates (Greiner) on a Genios plate reader (Tecan). The enzyme was combined with the peptide (inhibitor) in a buffer containing 100mM HEPES (pH 7.5), 50mM $CaCl_2$, 0.025% Tween-20, 5% DMSO and 1mM substrate. The rate of substrate hydrolysis was measured by monitoring the change in absorbance at 405 nm over 30 min to verify the linearity of the reaction curve. The average rate from minute 1 through minute 10 was used for all calculations. Initial calculations of background subtraction, average rate, duplicate averaging and % inhibition were made using the Magellan software (version 5) from Tecan. IC_{50} calculations were made using GraFit (version 5.0.10) from Erithacus Software by fitting inhibition data from 6 different inhibitor concentrations to the following 4-parameters equation (s = slope factor; x = inhibitor concentration; y = % inhibition at a given concentration of the inhibitor):

$$y = \frac{100\%}{1 + \left(\frac{x}{IC_{50}}\right)^{s}}$$

[0097] The K_m value for the serine protease substrate was determined from a Lineweaver-Burke plot (GraFit v5). The K_i value for the inhibitor was calculated using the formula $K_i = IC_{50} / (1+([substrate]/K_m))$. Increasing concentrations of the substrate were reacted with the enzyme, and the rate of each reaction (ABS/msec) was plotted against the substrate concentration. The reciprocal plot (Lineweaver-Burke) was also plotted to give K_m and V_{max} (inset) (A. J. Barrtt, Methods in Enzymology 1981, 80, 561-565; R. J. Leatherbarrow, 1992, GraFit, Erithacus Software Ltd., Staines, U.K.).

Table 2

Enzyme / Supplier	Enzyme concentration in assay	Substrate / Supplier	Substrate concentration in assay (mM)
Elastase from human neutrophils / Serva	0.6 mU/reaction	N-Met-Ala-Pro-Val-p-nitroanilide / Sigma	1
Cathepsin G from human neutrophils (CAS RN 107200-92-0) / Calbiochem	1 mU/reaction	N-Succinyl-Ala-Pro-Phe-p-nitroanilide / Sigma	1
Trypsin, iodination grade, from human pancreas (CAS RN 9002-07-7) / Calbiochem	1 mU/reaction	N-Benzoyl-Arg-p-nitroanilide / Sigma	0.32
Chymase from human skin / Calbiochem	9 mU/reaction	N-Succinyl-Ala-Pro-Phe-p-nitroanilide / Sigma	1.5
Thrombin from human plasma, high activity (CAS RN 9002-04-4) / Calbiochem	100 mU/reaction	Benzoyl-Phe-Val-Arg-p-nitroanilide / Calbiochem	0.5
Chymotrypsin from human pancreas (CAS RN 9004-07-3) / Calbiochem	1.6 microM/reaction	N-Succinyl-Ala-Pro-Phe-p-nitroanilide / Sigma	1
Coagulation factor Xa from human plasma (CAS RN 9002-05-5) / Calbiochem	0.4 mU/reaction	Methoxycarbonyl-D-Nle-Gly-Arg-p-nitroanilide / Roche	2
Tryptase from human lung / Calbiochem	12.5 mU/reaction	N-Benzoyl-Arg-p-nitroanilide / Sigma	1.28
Urokinase from human urine (CAS RN 9039-53-6) / Sigma Aldrich	250 mU/reaction	PyroGlu-Gly-Arg-p-nitroanilide x HCl / Endotell	0.5
Kallikrein from human plasma (CAS RN 9001-01-8) / Calbiochem	0.34 microg/reaction	N-Benzoyl-Pro-Phe-Arg-p-nitroanilide / Sigma	1
Plasmin from human plasma (CAS RN 9001-90-5) / Sigma-Aldrich	2 mU/reaction	D-Val-Leu-Lys-p-nitroanilide / Sigma	5

2.3. Cytotoxicity assay

[0098] The cytotoxicity of the peptide to HELA cells (Acc57) and COS-7 cells (CRL-1651) was determined using the MTT reduction assay (T. Mossman, J. Immunol. Meth. 1983, 65, 55-63; M. V. Berridge et al., Arch. Biochem. Biophys. 1993, 303, 474-482). HELA cells and COS-7 cells, respectively, were seeded at 7.0 x 10^3 and 4.5 x 10^3, respectively, cells per well and grown in 96-well microtiter plates for 24 h at 37°C and 5% CO_2. At this point, time zero (Tz) was determined by MTT reduction (see below). The supernatant of the remaining wells was discarded, and fresh medium and the peptide in serial dilutions of 12.5, 25 and 50 microM were pipetted into the wells. Each peptide concentration was assayed in triplicate. The incubation of the cells was continued for 48 h at 37°C and 5% CO_2. The wells were then washed once with phosphate buffered saline (PBS), and subsequently 100 microliters of MTT reagent (0.5 mg/ml in medium RPMI1640 and, respectively, DMEM) were added to the wells. It was incubated at 37°C for 2 h, and subsequently the medium was aspirated, and 100 microliters of isopropanol were added to each well. The absorbance at 595 nm of the solubilized product was measured (OD_{595}peptide). For each concentration, an average was calculated from triplicates. The percentage of growth was calculated using the formula (OD_{595}peptide-OD_{595}Tz-OD_{595}empty well) / (OD_{595}Tz-OD_{595}empty well) x 100% and plotted for each peptide concentration. The LC_{50} (the concentration, that kills 50% of the cells) value was determined for each peptide by using the trend line function of EXCEL (Microsoft Office 2000) for the concentrations (50, 25, 12.5 and 0 microM), the corresponding growth percentages and the value -50 (=TREND(C50:C0,%50:%0,-50)). The GI_{50} (growth inhibition) concentration was calculated for each peptide by using the trend line function for the concentrations (50, 25, 12.5 and 0 microg/ml), the corresponding percentages and the value 50 (=TREND(C50:C0,%50:%0,50)).

2.4. Hemolysis

[0099] The peptides were tested for their hemolytic activity against human red blood cells (hRBCs). Fresh hRBCs were washed three times with PBS by centrifugation for 10 min at 2000 x g. The peptide, at a concentration of 100 microM, was incubated with 20% (v/v) hRBCs for 1 h at 37°C. The final erythrocytes concentration was approximately 0.9 x 10^9 cells per ml. A value of 0% and, respectively, 100% cell lysis was determined by incubation of the hRBCs in the presence of PBS alone and, respectively, 0.1% Triton X-100 in H_2O. The sample was centrifuged, the supernatant was 20-fold diluted in PBS, and the optical density (OD) of the sample at 540 nm was measured. The 100% lysis value ($OD_{540}H_20$) gave an OD_{540} of approximately 1.3-1.8. The percentage of hemolysis was calculated using the formula (OD_{540}peptide / $OD_{540}H_20$) x100%.

2.5. Plasma stability

[0100] 405 microliters of plasma / albumin solution were placed into a polypropylene (PP) tube and spiked with 45 microliters of a 100 mM solution B, derived from 135 microliters of

PBS and 15 microliters of 1 mM peptide in PBS, pH 7.4. 150 microliters aliquots were transferred into individual wells of a 10 kDa filter plate (Millipore MAPPB 1010 Biomax membrane). For "0 minutes controls", 270 microliters of PBS were placed into a PP tube, and 30 microliters of the solution B were added and vortexed. 150 microliters of the control solution were placed into one well of the filter plate and served as "filtered control". A further 150 microliters of the control solution were placed directly into a receiver well (reserved for filtrate) and served as "not-filtered control". The entire plate, including the evaporation lid, was incubated for 60 min at 37°C. The plasma samples (rat plasma: Harlan Sera lab UK; human plasma: Blutspendezentrum Zürich) were centrifuged at least for 2 h at 4300 rpm (3500 g) and 15°C, in order to yield 100 microliters of filtrate. For "serum albumin" samples (freshly prepared human albumin: Sigma A-4327; rat albumin: Sigma A-6272; all at 40 mg/ml concentration in PBS), approximately 1 h of centrifugation was sufficient. The filtrates in the receiver plate were analysed by LC / MS [column: Jupiter C18 (Phenomenex); mobile phases: (A) 0.1% formic acid in water, (B) acetonitrile; gradient: 5% to 100% (B) in 2 min; electrospray ionization; MRM detection (triple quadrupole)]. The peak areas were determined, and triplicate values were averaged. The binding was expressed in percent of the (filtered and not-filtered time point 0 min) controls 1 and 2 by 100-(100 x T_{60}/T_0). The average from these values was then calculated.

2.6. Results

[0101] The results of the experiments described in sections 2.2. to 2.4. are indicated in table 3.

Table 3

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
1	86	>100000	92.6	7.8	0	1.1	5.7	5.7	0	nd	0
2	84	>100000	92	2.9	0	9.2	5.3	0.9	39.6	nd	nd
3	51	>100000	92	0	1	0	4	4	68	100	0
4	91	>100000	96	1.8	0	0	2.4	5.4	0	100	0
5	56	>100000	92	3	0	0.5	0.2	5.7	74	nd	0
6	?	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
9	105	4.1 at 100 microM (%)	88.1	nd	nd	nd	nd	nd	nd	nd	nd
10	75	0.3 at 100 microM (%)	89.9	nd	9.4	nd	nd	nd	nd	nd	nd
11	95	19 at 100 microM (%)	6.5	73.6	12.1	nd	nd	nd	nd	nd	nd
12	90	37038	97	28	12	11	5	12	59.3	59.3	nd
13	100	8.2 at 100 microM	95.0	nd	19.9	nd	nd	nd	nd	nd	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
		(%)									
14	52	>100000	88	0	42.3	8.7	6	5.4	84.2	100	0
15	56.0	>100000	95.0	54.2	12.7	nd	nd	nd	nd	100	0
16	66	>100000	90.0	17.9	12.9	nd	nd	3.2	nd	94	0.1
17	55	>100000	90.0	16	27.6	0	nd	nd	90.4	94	0.1
18	47	>100000	84	25	32.5	0	nd	nd	88.3	100	0
19	41	>100000	94.0	0	26.9	11	32	4	85.2	100	0
20	48	>100000	97.0	0	44.1	28	25	6.7	nd	100	0
21	97	16.4	95.6	2.6	5	nd	nd	nd	nd	nd	nd
22	55	>100000	84	0	98.8	nd	nd	5.7	3.8	8	0
23	38	>100000	90	4	60	0	11	9	29	51	0
24	71	>100000	97	1.0	1.2	3.5	30	5.1	0	99	nd
25	102	3.2 at 100 microM (%)	89.3	nd	10.0	nd	nd	nd	nd	nd	nd
26	49	>100000	84	2.2	0	3	6	3.1	66.4	nd	nd
27	48	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
28	39	>100000	95	32	0	12	6	1	0	nd	nd
29	42	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
30	39	49900	98	49	0	2	3	9	nd	nd	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
31	34	>100000	98	15	12	10	8	15	76	nd	nd
32	52	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
33	45	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
34	56	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
35	54	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
36	41	nd	nd	nd	nd	nd	nd	0	73.3	83	0
37	35	nd	nd	nd	nd	nd	nd	5	56	92	0.1
38	31	>100000	96	4	1	0	0	1	11	100	0
39	38	>100000	94	7	0	2	0	2	34	98	0
40	25	>44862	94	19	8	1	3	10	33	97	0.1
41	49	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
42	46	nd	nd	nd	nd	nd	nd	7	0	87	0
43	77	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
44	31	>10000	100	24	3	9	9	14	50	67	0.1
45	47	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
52	52	>100000	86	0	42.3	8.7	6	5.4	84.2	100	0
53	56	>100000	95	54	12.7	nd	nd	nd	nd	63	0
54	66	>100000	90	18	12.9	nd	nd	3.2	nd	nd	nd
55	55	>100000	90	16	27.6	nd	nd	nd	90.4	94	0.1
56	47	>100000	84	25	32.5	0	nd	nd	88.3	100	0

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
57	41	>100000	94	0	26.9	11	32	4	82.2	0	0
58	47.5	>100000	97	0	44.1	28	25	6.7	nd	100	0
59	55	>100000	84	0	98.8	nd	nd	5.7	3.8	8	0
60	38	>100000	90	4	60	0	11	9	29.4	51	0
61	72	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
62	69	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
63	41	>100000	96	11	7	1	0	0	50	87	0
65	47	nd	nd	nd	nd	nd	nd	1	57	84	0
66	48	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
67	48	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
70	69	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
71	70	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
76	nd	71	nd	nd	nd	nd	nd	nd	nd	nd	nd
77	nd	68	nd	nd	nd	nd	nd	nd	nd	nd	nd
78	nd	29	nd	<4000	nd	nd	nd	nd	nd	61	nd
79	nd	66	nd	nd	nd	nd	nd	nd	nd	nd	nd
80	nd	35	nd	>20000	nd	nd	nd	nd	nd	12	nd
81	61.3 at 100 microM (%)	28	100000	100000	nd	13.1	8.7	nd	nd	58	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
82	nd	18	nd	72.9	nd	nd	15.3	nd	44.5	nd	nd
83	nd	43	nd	100000	nd	nd	nd	nd	nd	12	nd
84	20195	18	10.8	17103	0	20.6	13.3	10.4	4.2	9	nd
85	nd	28	0	>20000	nd	12.6	25.6	nd	nd	nd	nd
86	47 at 100 microM (%)	26	0	>100000	0	10.7	24.8	nd	0	nd	nd
87	nd	37	nd	106977	nd	nd	nd	nd	nd	65	nd
88	>100000	18	6.4	4309	0	0.2	3	96	0.6	73	nd
89	nd	43	nd	nd	nd	nd	nd	nd	nd	51	nd
90	66975	21	5.2	33074	0	0	5.5	3.5	5	96	nd
91	45 at 100 microM (%)	28	0	48108	4.7	13.5	19.4	nd	nd	79	nd
92	nd	43	nd	nd	nd	nd	nd	nd	nd	93	nd
93	nd	41	nd	nd	nd	nd	nd	nd	5.6	100	nd
94	nd	50	nd	nd	nd	nd	nd	nd	nd	nd	nd
95	38677	24	8.9	33729	0	0	11.3	10.3	0	89	nd
96	21175	15	7.5	15433	0	3.6	0	6.2	0	52	nd
97	>100000	24	9.5	77431	0	11.6	4.8	11.9	0	100	nd
98	>100000	21	0	38820	0	5.2	0	0	0	78	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
99	85196	16	30.5	8558	0	0	0	17.4	0	58	nd
100	nd	35	nd	nd	nd	nd	nd	nd	nd	83	nd
101	nd	49	nd	nd	nd	nd	nd	nd	nd	nd	nd
102	>100000	13	0	4975	0	1.7	0	0.5	0	55	nd
103	>100000	18	6.4	4309	0	10.2	3	9.6	0.6	47	nd
104	53.5 at 100 microM (%)	34	0	3.1 at 100 microM (%)	0	7.7	6.2	0	0	nd	nd
105	nd	34	nd	nd	nd	nd	nd	nd	nd	nd	nd
106	nd	49	nd	nd	nd	nd	nd	nd	nd	nd	nd
107	nd	51	nd	nd	nd	nd	nd	nd	nd	nd	nd
108	nd	31	nd	nd	nd	nd	nd	nd	nd	nd	nd
109	54.1 at 100 microM (%)	33	0	13.8	0.1	0	5.6	nd	nd	nd	nd
110	nd	38	nd	nd	nd	nd	nd	nd	nd	nd	nd
111	nd	46	nd	nd	nd	nd	nd	nd	nd	nd	nd
112	nd	39	nd	nd	nd	nd	nd	nd	nd	33	nd
113	nd	35	nd	nd	nd	nd	nd	nd	nd	nd	nd
114	nd	47	nd	nd	nd	nd	nd	nd	nd	34	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
129	nd	30	nd	nd	nd	nd	nd	nd	nd	nd	nd
131	45	28	0	nd	46108	nd	nd	nd	nd	nd	nd
132	nd	26	nd	nd	nd	nd	nd	nd	nd	nd	nd
133	nd	26	nd	nd	nd	nd	nd	nd	nd	nd	nd
134	nd	23	nd	nd	nd	nd	nd	nd	nd	nd	nd
135	nd	23	nd	nd	nd	nd	nd	nd	nd	nd	nd
136	>100000	21	0	67.9	0	5.2	0	0	0	nd	nd
137	66975	21	5.2	68.7	0	0	5.5	3.5	5	nd	nd
138	43856	19	12.2	77.1	4.6	17.1	12.6	14.4	0	nd	nd
139	nd	18	nd	nd	nd	nd	nd	nd	nd	nd	nd
140	20195	18	10.8	79.6	0	20.6	13.3	10.4	4.2	nd	nd
141	63.4 at 100 microM (%)	18	0	72.9	0	0	15.3	nd	44.5	56	nd
142	nd	16	nd	nd	nd	nd	nd	nd	nd	nd	nd
143	28 at 100 microM (%)	15	12	91	0	12	0	8	18	nd	nd
144	21175	7.5	7.5	80.6	0	3.6	0	6.2	0	nd	nd
145	nd	14	nd	nd	nd	nd	nd	nd	nd	nd	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
146	1 at 100 microM (%)	12	3	87	0	11	1	0	22	nd	nd
147	nd	11	nd	nd	nd	nd	nd	nd	nd	nd	nd
148	52 at 100 microM (%)	11	9	91	7	32	8	12	30	nd	nd
149	nd	11	nd	nd	nd	nd	nd	nd	nd	nd	nd
150	nd	10	nd	nd	nd	nd	nd	nd	nd	nd	nd
151	nd	10	nd	nd	nd	nd	nd	nd	nd	nd	nd
152	nd	9	nd	nd	nd	nd	nd	nd	nd	nd	nd
153	56 at 100 microM (%)	8.5	8	84	0	16	11	16	9	nd	nd
154	27 at 100 microM (%)	8.3	0	4	0	7	0	1	15	nd	nd
155	52 at 100 microM (%)	8.2	18	83	3	19	9	12	30	nd	nd
156	46 at 100 microM (%)	7.5	0	5	0	17	0	7	15	nd	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
157	nd	7	nd	nd	nd	nd	nd	nd	nd	nd	nd
158	55 at 100 microM (%)	7.1	8	93	0	2	1	10	13	nd	nd
159	nd	7	nd	nd	nd	nd	nd	nd	nd	nd	nd
160	55 at 100 microM (%)	6	3	94	2	23	1	14	30	nd	nd
161	nd	6	nd	nd	nd	nd	nd	nd	nd	nd	nd
162	nd	12.5	nd	nd	nd	nd	nd	nd	nd	nd	nd
179	nd	50	>100000 [IC_{50} (nmol)]	76.0	nd	nd	nd	nd	0	nd	nd
180	120	nd	60	nd	nd	nd	nd	nd	<100	nd	nd
181	127	nd	113	nd	nd	nd	nd	nd	40	nd	nd
182	111	nd	59	nd	nd	nd	nd	nd	39	nd	nd
184	221	nd	48	nd	nd	nd	nd	nd	27	nd	nd
185	514	nd	126	nd	nd	nd	nd	nd	14	nd	nd
186	337	nd	99	nd	nd	nd	nd	nd	15	nd	nd
187	158	nd	39	nd	nd	nd	nd	nd	<100	nd	nd
188	105	nd	34	nd	nd	nd	nd	nd	<100	nd	nd
189	164	nd	39	nd	nd	nd	nd	nd	<100	nd	nd

Example	Cathepsin G IC_{50} (nmol)	Elastase IC_{50} (nmol)	Trypsin at 100 microM (%)	Chymotrypsin at 100 microM (%)	Chymase at 100 microM (%)	Thrombin at 100 microM (%)	Factor Xa at 100 microM (%)	Urokinase at 100 microM (%)	Tryptase at 100 microM (%)	Cytotoxicity LC_{50} / GI_{50} Hela cells	Hemolysis at 100 microM (%)
191	400	nd	66	nd	nd	nd	nd	nd	21	nd	nd
192	650	nd	72	nd	nd	nd	nd	nd	16	nd	nd
193	431	nd	35	nd	nd	nd	nd	nd	6	nd	nd
194	1570	nd	431	nd	nd	nd	nd	nd	9	nd	nd
195	4000	nd	108	nd	nd	nd	nd	nd	12	nd	nd
nd: Not determined											

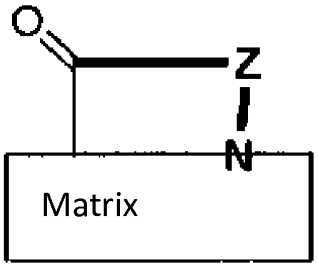
[0102] The results of the experiments described in section 2.5. are indicated in table 4.

Table 4

Example	Stability Human Plasma $t_{1/2}$ (min)	Stability Rat Plasma $t_{1/2}$ (min)
22	300	300
23	300	300
158	300	300

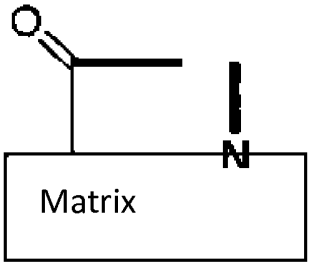
Patentkrav

1. Forbindelse med formlen



(I),

i hvilken



er en dipeptidrest af to forskellige aminosyrebyggesten, hvor dipeptidet er ^{D}Pro-^{L}Pro, og

Z er en undecapeptidkæde af elleve aminosyrerester, hvor P1-resten er resten af Nle, Ile, Aoc, hLeu, Chg, OctG, hPhe, 4AmPhe, Cha, Phe, Tyr, 2Cl-Phe, Trp, 1-Nal eller Leu;

P2-resten er resten af Cys, Glu, Nle, Thr eller Gln;

P3-resten er resten af Thr, Ala eller Abu;

P4-resten er resten af Lys, Nle, Ala, Abu eller Thr;

P5-resten er resten af Ser, AlloThr eller Dpr;

P6-resten er resten af Ile, C_5al, Leu, Nle, Aoc, OctG, Cha, hLeu, hPhe, Chg, t-BuA, Glu eller Asp;

P7-resten er resten af Pro;

P8-resten er resten af Pro, Ala eller Pro(4NHCOPhe);

P9-resten er resten af Tyr, Phe, Ile, Nle, Cha, Gln, Arg, Lys, His eller Thr;

P10-resten er resten af Cys, Arg, Nle, Gln, Lys, Met, Thr eller Ser; og P11-resten er resten af Tyr, Gln, Arg, Ser, Nle, 2-Nal, 2Cl-Phe, Cha, Phg, Phe, Asp, Asn eller Thr,

to rester af Cys, som foreligger som P2- og P10-resterne, der er forbundet ved hjælp af en disulfidbro dannet ved erstatning af de to -SH-grupper i de to rester af Cys med en -S-S-gruppe,
i fri form eller i en farmaceutisk acceptabel saltform.

2. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Phe, hPhe, 4AmPhe, Nle, Chg, Ile, Tyr, Trp, 2Cl-Phe, 1-Nal eller Cha;
P2-resten er resten af Cys, Glu eller Nle;
P3-resten er resten af Thr;
P4-resten er resten af Lys eller Nle;
P5-resten er resten af Ser, AlloThr eller Dpr;
P6-resten er resten af Asp eller Glu;
P8-resten er resten af Pro;
P9-resten er resten af Ile, Nle, Cha, Gln eller Tyr;
P10-resten er resten af Cys, Arg eller Nle; og
P11-resten er resten af Thr, Asp, Ser, Tyr, Phe, Asn eller Arg.

3. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Ile, Nle, Aoc, hLeu, Chg, OctG eller hPhe;
P2-resten er resten af Cys, Glu, Thr eller Gln;
P3-resten er resten af Thr, Ala eller Abu;
P4-resten er resten af Ala, Thr eller Abu;
P5-resten er resten af Ser;
P6-resten er resten af OctG, Ile, Cha, Leu, C_5al, Nle, Aoc, Chg, t-BuA eller hLeu;
P8-resten er resten af Pro eller Pro(4NHCOPhe);
P9-resten er resten af Gln, Tyr, Ile eller Phe;
P10-resten er resten af Cys, Lys, Gln, Thr, Met eller Arg; og
P11-resten er resten af Tyr, Ser, Arg, Gln, Nle, 2-Nal, 2Cl-Phe, Phe, Cha eller Phg.

4. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Cha, Tyr eller Trp;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Leu;
P8-resten er resten af Pro;
P9-resten er resten af Lys;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

5. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Phe;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;
P9-resten er resten af Ile;
P10-resten er resten af Cys; og
P11-resten er resten af Tyr.

6. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Ile;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;

P9-resten er resten af Nle;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

7. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Ile;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;
P9-resten er resten af Cha;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

8. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Ile;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Nle;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;
P9-resten er resten af Ile;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

9. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Phe;
P2-resten er resten af Cys;
P3-resten er resten af Thr;

P4-resten er resten af Nle;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;
P9-resten er resten af Ile;
P10-resten er resten af Cys; og
P11-resten er resten af Tyr.

10. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Chg;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;
P9-resten er resten af Ile;
P10-resten er resten af Cys; og
P11-resten er resten af Tyr.

11. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Nle;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Ile;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Gln.

12. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Nle;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Ile;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Tyr.

13. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af hPhe;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af OctG;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Gln.

14. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Gln;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Ile;
P8-resten er resten af Pro;

P9-resten er resten af Gln;
P10-resten er resten af Thr; og
P11-resten er resten af Tyr.

15. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af hPhe;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Cha;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Phe.

16. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Glu;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Ile;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Lys; og
P11-resten er resten af Tyr.

17. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Cys;
P3-resten er resten af Thr;

P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Cha;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Phe.

18. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af hPhe;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Cha;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Gln.

19. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Cha;
P8-resten er resten af Pro(4NHCOPhe);
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Gln.

20. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af hPhe;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af OctG;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Tyr.

21. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Chg;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;
P9-resten er resten af Ile;
P10-resten er resten af Cys; og
P11-resten er resten af Thr.

22. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Chg;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Asp;
P8-resten er resten af Pro;

P9-resten er resten af Ile;
P10-resten er resten af Cys; og
P11-resten er resten af Asp.

23. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Cha;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Gin.

24. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af Ile;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Tyr.

25. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af OctG;
P2-resten er resten af Cys;
P3-resten er resten af Thr;

P4-resten er resten af Ala;
P5-resten er resten af Ser;
P6-resten er resten af OctG;
P8-resten er resten af Pro;
P9-resten er resten af Gln;
P10-resten er resten af Cys; og
P11-resten er resten af Gln.

26. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Cha;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Leu;
P8-resten er resten af Pro;
P9-resten er resten af Lys;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

27. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Tyr;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Leu;
P8-resten er resten af Pro;
P9-resten er resten af Lys;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

28. Forbindelse ifølge krav 1 med formlen I, i hvilken, i undecapeptidkæden,
P1-resten er resten af Trp;
P2-resten er resten af Cys;
P3-resten er resten af Thr;
P4-resten er resten af Lys;
P5-resten er resten af Ser;
P6-resten er resten af Leu;
P8-resten er resten af Pro;
P9-resten er resten af Lys;
P10-resten er resten af Cys; og
P11-resten er resten af Arg.

29. Forbindelse ifølge krav 1 med formlen I i form af en enantiomer.

30. Forbindelse som defineret i et af kravene 1 til 29 med formlen I til anvendelse som en terapeutisk aktiv substans.

31. Forbindelse ifølge krav 30 med formlen I med selektiv protease hæmmende aktivitet, især mod cathepsin G, elastase eller tryptase; og/eller med anticeraktivitet; og/eller med antiinflammatorisk aktivitet; og/eller med antiinfektiøs aktivitet; og/eller med aktivitet mod en kardiovaskulær sygdom; og/eller med aktivitet mod en immunologisk sygdom; og/eller med antineurodegenerativ aktivitet; og/eller med aktivitet mod en lungesygdom.

32. Farmaceutisk sammensætning omfattende en forbindelse som defineret i et af kravene 1 til 29 med formlen I som aktiv bestanddel og en farmaceutisk acceptabel bærer.

33. Sammensætning ifølge krav 32, **kendetegnet ved,** at den foreligger i en form, der er egnet til oral, bukkal, rektal, vaginal, topisk, transdermal, transmucosal, pulmonal indgivelse eller indgivelse ved injektion, inhalation eller implantation.

34. Sammensætning ifølge krav 32 eller krav 33, **kendetegnet ved,** at den foreligger i form af en tablet, en dragé, en kapsel, en tablet, en pille, et pulver, en væske, en opløsning, en sirup, en eliksir, en opslæmning, en suspension, en emulsion, en gel, en creme, en salve, et plaster, en spray, en forstøver, en inhalator, en insufflator, et suppositorium, et system med forsinket frigivelse, en langtidsvirkende formulering, et depotpræparat eller et liposom.

35. Anvendelse af en forbindelse som defineret i et af kravene 1 til 29 med formlen I til fremstilling af et medikament til inhibering af et proteaseenzym.

36. Anvendelse ifølge krav 35, **kendetegnet ved,** at medikamentet er beregnet til at blive anvendt til forebyggelse af en infektion i et rask individ eller til at sinke en infektion i en inficeret patient; eller hvor cancer er medieret af eller resulterer af proteaseaktivitet; eller hvor en immunologisk sygdom er medieret af eller resulterer af proteaseaktivitet; eller hvor en inflammation er medieret af eller resulterer af proteaseaktivitet; eller hvor en immunologisk reaktion er medieret af eller resulterer af proteaseaktivitet.

37. Anvendelse af en forbindelse som defineret i et af kravene 2 og 5 til 10 med formlen I til fremstilling af et medikament til inhibering af cathepsin G.

38. Anvendelse af en forbindelse som defineret i et af kravene 3 og 11 til 22 med formlen I til fremstilling af et medikament til inhibering af elastase.

39. Anvendelse af en forbindelse som defineret i et af kravene 4 og 23 til 25 med formlen I til fremstilling af et medikament til inhibering af tryptase.

40. Fremgangsmåde til fremstilling af en forbindelse som defineret i et af kravene 1 til 28 med formlen I, hvilken fremgangsmåde omfatter
(a) at forbinde en passende funktionaliseret fast bærer med et passende N-beskyttet derivat af den aminosyre, hvilken i det ønskede slutprodukt er i po-

sition 5, 6 eller 7, hvor enhver funktionel gruppe, som foreligger i det N-beskyttede aminosyrederivat, ligeledes er passende beskyttet;

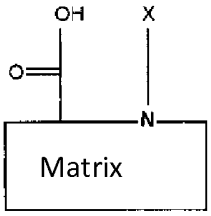
(b) at fjerne den N-beskyttende gruppe fra det således opnåede produkt;

(c) at forbinde det således opnåede produkt med et passende N-beskyttet derivat af den aminosyre, hvilken i det ønskede slutprodukt er en position nærmere den N-terminale aminosyrerest, hvor enhver funktionel gruppe, som foreligger i det N-beskyttede aminosyrederivat, ligeledes er passende beskyttet;

(d) at fjerne den N-beskyttende gruppe fra det således opnåede produkt;

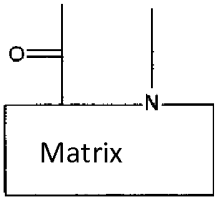
(e) at gentage trin (c) og (d), indtil den N-terminale aminosyrerest er blevet indført;

(f) at forbinde det således opnåede produkt med en forbindelse med formlen



(II),

i hvilken



er som defineret i krav 1, og X er en N-beskyttende gruppe eller, alternativt,

(fa) at forbinde produktet opnået i trin (e) med et passende N-beskyttet derivat af aminosyren L-prolin;

(fb) at fjerne den N-beskyttende gruppe fra det således opnåede produkt; og

(fc) at forbinde det således opnåede produkt med et passende N-beskyttet derivat af aminosyren D-prolin;

(g) at fjerne den N-beskyttende gruppe fra produktet opnået i trin (f) eller trin (fc);

(h) at forbinde det således opnåede produkt med et passende N-beskyttet derivat af den aminosyre, hvilken i det ønskede slutprodukt er i position 11, hvor enhver funktionel gruppe, som foreligger i det N-beskyttede aminosyrederivat, ligeledes er passende beskyttet;

(i) at fjerne den N-beskyttende gruppe fra det således opnåede produkt;

(j) at forbinde det således opnåede produkt med et passende N-beskyttet derivat af den aminosyre, hvilken i det ønskede slutprodukt er en position længere væk fra position 11, hvor enhver funktionel gruppe, som foreligger i det N-beskyttede aminosyrederivat, ligeledes er passende beskyttet;

(k) at fjerne den N-beskyttende gruppe fra det således opnåede produkt;

(l) at gentage trin (j) og (k), indtil alle aminosyrerester er blevet indført;

(m) hvis det ønskes, selektivt at afbeskytte en eller flere beskyttet/beskyttede funktionel(le) gruppe(r), der foreligger i molekylet, og passende at substituere den/de reaktive gruppe(r), der således er frigivet;

(n) hvis det ønskes, at danne en interstreng-binding mellem sidekæder af passende aminosyrerester ved position 2 og 10;

(o) at løsne det således opnåede produkt fra den faste bærer;

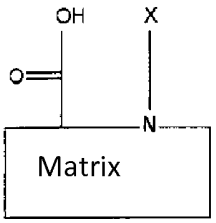
(p) at cyklisere det spaltede produkt fra den faste bærer;

(q) at fjerne enhver beskyttende gruppe, der foreligger på en funktionel gruppe af enhver aminosyrerest i det således opnåede cykliske produkt og, hvis det ønskes, enhver beskyttende gruppe, som derudover foreligger i det således opnåede molekyle; og

(r) hvis det ønskes, at omdanne en således opnået forbindelse med formlen I i fri form til et farmaceutisk acceptabelt salt eller at omdanne et således opnået farmaceutisk acceptabelt eller uacceptabelt salt til den tilsvarende forbindelse med formlen I i fri form eller til et andet, farmaceutisk acceptabelt, salt.

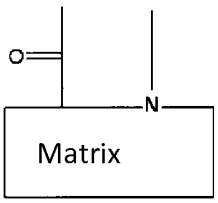
41. Fremgangsmåde til fremstilling af en forbindelse som defineret i et af kravene 1 til 28 med formlen I, hvilken fremgangsmåde omfatter

(a') at forbinde en passende funktionaliseret fast bærer med en forbindelse med formlen



(II),

i hvilken



er som defineret i krav 1, og X er en N-beskyttende gruppe eller, alternativt,

(a'a) at forbinde den passende funktionaliserede faste bærer med et passende N-beskyttet derivat af aminosyren L-prolin;

(a'b) at fjerne den N-beskyttende gruppe fra det således opnåede produkt; og

(a'c) at forbinde det således opnåede produkt med et passende N-beskyttende derivat af aminosyren D-prolin;

(b') at fjerne den N-beskyttende gruppe fra produktet opnået i trin (a') eller trin (a'c);

(c') at forbinde det således opnåede produkt med et passende N-beskyttet derivat af den aminosyre, hvilken i det ønskede slutprodukt er i position 11, hvor enhver funktionel gruppe, som foreligger i det N-beskyttede aminosyrederivat, ligeledes er passende beskyttet;

(d') at fjerne den N-beskyttende gruppe fra det således opnåede produkt;

(e') at forbinde det således opnåede produkt med et passende N-beskyttet derivat af den aminosyre, hvilken i det ønskede slutprodukt er en position længere væk fra position 11, hvor enhver funktionel gruppe, som foreligger i det N-beskyttede aminosyrederivat, ligeledes er passende beskyttet;

(f') at fjerne den N-beskyttende gruppe fra det således opnåede produkt;

(g') at gentage trin (e') og (f'), indtil alle aminosyrerester er blevet indført;

(h') hvis det ønskes, selektivt at afbeskytte en eller flere beskyttet/beskyttede funktionel(le) gruppe(r), der foreligger i molekylet, og passende at substituere den/de reaktive gruppe(r), der således er frigivet;

(i') hvis det ønskes, at danne en interstreng-binding mellem sidekæder af passende aminosyrerester ved position 2 og 10;

(j') at løsne det således opnåede produkt fra den faste bærer;

(k') at cyklisere det spaltede produkt fra den faste bærer;

(l') at fjerne enhver beskyttende gruppe, der foreligger på en funktionel gruppe af enhver aminosyrerest i det således opnåede cykliske produkt og, hvis det ønskes, enhver beskyttende gruppe, som derudover foreligger i det således opnåede molekyle; og

(m') hvis det ønskes, at omdanne en således opnået forbindelse med formlen I i fri form til et farmaceutisk acceptabelt salt eller at omdanne et således opnået farmaceutisk acceptabelt eller uacceptabelt salt til den tilsvarende forbindelse med formlen I i fri form eller til et andet, farmaceutisk acceptabelt, salt.

42. Fremgangsmåde ifølge krav 40 eller krav 41 til fremstilling af en forbindelse som defineret i krav 29 med formlen I, hvilken fremgangsmåde omfatter at anvende et chiralt udgangsmateriale i form af en enantiomer.