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Alfa-synuclein epitopok mimotópjainak alkalmazása Lewy-test betegségek kezelésében

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(54) **USE OF MIMOTOPES OF ALPHA-SYNUCLEIN EPITOPES FOR TREATING LEWY BODY DISEASES**

VERWENDUNG VON MIMOTOPEN VON ALPHA-SYNUCLEIN-EPITOPEN ZUR BEHANDLUNG VON LEWY-KÖRPER-ERKRANKUNGEN

UTILISATION DE MIMOTOPES D'ÉPITOPES D'ALPHA-SYNUCLÉINE POUR TRAITER DES MALADIES À CORPS DE LEWY

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Description

[0001] The present invention relates to a medicament to be used to prevent and/or treat synucleinopathies.

[0002] Synucleinopathies are a diverse group of neurodegenerative disorders that share a common pathologic characteristic: in neuropathologic examinations characteristic lesions can be detected containing abnormal aggregates of alpha-synuclein (alpha-syn, asyn) protein in selected populations of neurons and glia cells.

[0003] Alpha-syn (initially identified as PARK1 and PARK4) is a 140 amino acid protein widely expressed in the neocortex, hippocampus, dentate gyrus, olfactory bulb, striatum, thalamus and cerebellum. Alpha-Syn is also highly expressed in hematopoietic cells including B-, T-, and NK cells as well as monocytes and platelets. The exact role in these cells is not known but it has been implicated in the differentiation of megakaryocytes (platelet precursors).

[0004] The most common synucleinopathies include but are not limited to Lewy body disorders (LBDs) like Parkinson's disease (PD), Parkinson's disease with dementia (PDD) and dementia with Lewy bodies (DLB), as well as Multiple System Atrophy (MSA) or Neurodegeneration with Brain Iron Accumulation type I (NBIA Type I). The current treatment options for these diseases include symptomatic medications such as L-dopa, anticholinergic drugs as well as inhibitors of monoamine oxidase. However, all treatment opportunities currently present only lead to symptomatic alleviation but do not induce a long lasting, disease modifying effect in patients.

[0005] Lewy body disorders (LBD) are progressive neurodegenerative disorders characterized by tremor, rigidity, bradykinesia and by loss of dopaminergic neurons in the brain. In the case of DLB and PDD signs also include cognitive impairment. Up to 2% of the population above 60 years of age in western countries develop the typical signs of PD/LBD. Currently only symptomatic treatment is available. Unfortunately, these therapies only provide temporary relief from early symptoms and do not halt disease progression. The pathogenesis of PD/LBD is still incompletely understood, but it appears that genetic susceptibility and environmental factors are involved in the development of the disease. Despite all genetic advances, PD/LBD is primarily a sporadic disorder with no known cause (also called idiopathic PD/LBD).

[0006] Patients suffering from this disease develop characteristic ubiquitinated intracellular inclusions called Lewy bodies (LBs) in the cortical and subcortical areas of the brain. Especially regions with high content of dopaminergic neurons or neuronal projections show this typical pathologic feature. Recently, several studies could show that the synaptic protein alpha-syn plays a central role in LBD pathogenesis. In LBD, alpha-syn accumulates in LBs throughout affected brain areas. Additionally, it could be demonstrated that single point mutations as well as duplications or multiplications in the alpha-syn gene are associated with rare familial forms of parkinsonism. Importantly, based on results from overexpression studies in transgenic (tg) mice as well as in *Drosophila melanogaster* its key role in the pathogenesis of PD/LBD is underscored as these animal models mimic several characteristics of PD.

[0007] Another very important synucleinopathy is Multiple System Atrophy (MSA). MSA is a sporadic neurodegenerative disorder that is characterised by symptoms of L-DOPA-resistant parkinsonism, cerebellar ataxia, and dysautonomia. Patients suffer from multi-system neuronal loss affecting various brain areas including striatum, substantia nigra, cerebellum, pons, as well as the inferior olives and the spinal cord. MSA is characterized by alpha-syn-positive glial cytoplasmic (GCI) and rare neuronal inclusions throughout the central nervous system. These inclusions are associated with striatonigral degeneration, olivopontocerebellar atrophy, and involvement of autonomic nuclei in medulla and spinal cord. The importance of GCIs for the pathogenesis of MSA is generally acknowledged and underscored by recent analysis of transgenic mouse models analysing the effect of alpha-syn overexpression in oligodendroglia. In tg mice overexpressing human alpha-syn both GCI-like aggregates and biochemical markers of MSA were observed.

[0008] Although the exact mechanisms by which accumulation of alpha-syn leads to the typical features of neurodegeneration in synucleopathies are not fully understood, recent studies imply that abnormal formation and accumulation of alpha-syn is involved in the degenerative processes underlying synucleinopathy. Recently, different forms of alpha-syn have been identified in LBs. Beside the full length form of the protein, different forms of modified alpha-syn have been identified including phosphorylated, nitrated, and mono-, di-, or tri-ubiquitinated alpha-syn. In addition, C-terminally truncated forms of the protein, like alpha-syn 1-119, alpha-syn 1-122 and alpha-syn 1-123, have been detected in brain tissue from both transgenic mice and PD cases. It is currently believed that up to 15% of the alpha-syn detected in LBs and lewy neurites is truncated. Previous in vitro studies using truncated alpha-syn could demonstrate that alpha-syn lacking the C-terminal 20-30 amino acids was showing an increased tendency to aggregate and to form filaments found in Lewy-neurites and LBs. These truncated versions could thus act in a similar way as truncated and modified forms of amyloid beta (A β) in Alzheimer's disease (AD). These truncated and modified forms of A β are thought to act as seed molecules for plaque deposition and show a higher aggregation propensity as well as high neurotoxicity and synaptotoxicity in vivo and in vitro.

[0009] Thus full length alpha-syn as well as truncated and/or modified forms of alpha-syn, which are showing potential seeding effects, are then believed to accumulate leading to oligomer-formation. Based on recent studies it is believed that such oligomer-formation for example in the synaptic terminals and axons plays an important role for PD/LBD development and could thus be enhanced by the presence of truncated forms of alpha-syn. Hence, reduction of alpha-syn deposition and oligomerisation should be beneficial in the treatment of synucleopathies, especially of idiopathic LBD/PD

and MSA and could present the first strategy for treatment of these neurodegenerative diseases in addition to the mere alleviation of symptoms resulting from current treatment strategies like L-DOPA application.

[0010] In Iwatsubo T. (Neuropathology 27 (5)(2007): 474-478) the correlation of alpha-synuclein depositions as well as its phosphorylation with a pathogenesis of alpha-synucleinopathies is examined. The author of this publication found that serine 129 of alpha-synuclein deposited in synucleopathy lesions is extensively phosphorylated. US 2007/213253 relates to mutant human alpha-synuclein as well as peptides derived therefrom which may be used for inhibiting the aggregation of the wild-type human alpha-synuclein. In the WO 2004/041067 means and methods for preventing or treating diseases associated with alpha-synuclein aggregation are disclosed which comprise the use of alpha-synuclein fragments. In the US 2003/166558 peptides are described which can be used to induce immune response to protein deposits. US 2005/198694 relates to alpha-synuclein fragments comprising at least 100 amino acids and having a C-terminal deletion of 1 to 23 amino acids.

[0011] Liang et al. (J. Neurochem. 99(2006): 470-482) studied the regulation of alpha-synuclein in rats. They observed that in alcohol preferring rats the expression rate of alpha-synuclein is increased compared to alcohol-non preferring rats.

[0012] In Hamilton BA (Genomics 83(2004): 739-742) the distribution of alpha-synuclein 53Thr and 53Ala in primates is examined.

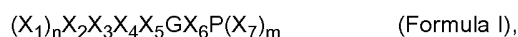
[0013] In WO 2004/041067 immunogenic alpha-synuclein fragments are disclosed which are able to induce an immune response against a specific epitope within residues 70-140 of alpha-synuclein.

[0014] WO 2006/045037 relates to C-terminal truncated alpha-synuclein molecules which can be used to screen for agents which have a pharmacological activity useful for treating a Lewy Body Disease.

[0015] Although experimental therapies utilizing neurotrophic factors and grafting of dopaminergic cells have yielded promising results, alternative approaches designed to reduce the neuronal accumulation of alpha-syn are required. There is compelling evidence accumulating that alpha-syn aggregates might be targeted by immunotherapy. Indeed, recently a potential for the treatment of synucleinopathies has been shown. Tg mice overexpressing human alpha-syn were vaccinated with human alpha-syn protein. In mice that produced high relative affinity antibodies upon vaccination, there was decreased accumulation of aggregated alpha-syn in neuronal cell bodies and synapses which was associated with reduced neurodegeneration. Furthermore, antibodies produced by immunized animals also detected abnormal aggregated forms of alpha-syn associated with the neuronal membrane and promoted the degradation of these aggregates, probably via lysosomal pathways. Similar effects were observed using passive immunotherapy with an exogenously applied alpha-syn-specific antibody. These results suggest that vaccination is effective in reducing neuronal accumulation of alpha-syn aggregates and that further development of this approach might elicit beneficial effects in the treatment of LBD and synucleinopathies.

[0016] It is an object of the present invention to provide a medicament to prevent and treat synucleinopathies on the basis of a vaccine.

[0017] The present invention relates to the provision of at least one peptide or polypeptide comprising the amino acid sequence



wherein

X_1 is any amino acid residue,

X_2 is an amino acid residue selected from the group consisting of lysine (K), arginine (R), alanine (A) and histidine (H),

X_3 is an amino acid residue selected from the group consisting of asparagine (N), glutamine (Q), serine (S), glycine (G) and alanine (A), preferably asparagine (N), serine (S), glycine (G) and alanine (A),

X_4 is an amino acid residue selected from the group consisting of glutamic acid (E), aspartic acid (D) and alanine (A), X_5 is an amino acid residue selected from the group consisting of glutamic acid (E) and aspartic acid (D),

X_6 is an amino acid residue selected from the group consisting of alanine (A) and tyrosine (Y),

X_7 is any amino acid residue,

n and m , independently, are 0 or an integer of more than 0,

and wherein the amino acid sequence according to Formula I is not identical with, or does not comprise the 7-mer polypeptide fragment of alpha-synuclein having the amino acid sequence KNEEGAP,

said at least one peptide or polypeptide having a binding capacity to an antibody which is specific for an epitope of alpha-synuclein comprising the amino acid sequence KNEEGAP,

for use in preventing and/or treating synucleinopathies.

[0018] These peptides or polypeptides according to the present invention can be provided in compositions suitable for the intended use for preventing and/or treating synucleinopathies, especially in pharmaceutical compositions, preferably combined with a pharmaceutically acceptable carrier. Such pharmaceutical compositions can be administered

to a patient in need thereof in an effective amount to achieve the preventive and/or therapeutic effect.

[0019] The peptides and polypeptide according to the present invention are able to induce the *in vivo* formation of antibodies directed (binding) to alpha-synuclein and fragments thereof, in particular to fragments of alpha-synuclein comprising the amino acid sequence KNEEGAP. Antibodies directed (binding) to said peptides and polypeptides, however, show no or substantially no immune reactivity to beta-synuclein (beta-syn, b-syn). Therefore, unlike the original alpha-synuclein or fragment(s) thereof, the peptides and polypeptides according to the present invention provide a specificity towards the disease related agent and avoid cross reactivity with disease unrelated synucleins. This strongly suggests significant superiority regarding efficacy and safety, the latter in particular because of the neuroprotective characteristics that have been described for beta-synuclein (Hashimoto M. et al., J Biol Chem. 2004 May 28;279(22):23622-9. Hashimoto M, Neuron. 2001 Oct 25;32(2):213-23).

[0020] The alpha-synuclein specific antibodies induced by the administration of the compounds of the present invention might not only bind to monomeric forms of alpha-synuclein but also to multimeric forms. This allows to reduce the amount of oligomers of alpha-synuclein in the body of an individual to be treated. The reduction of alpha-synuclein is particularly beneficial in the treatment of synucleopathies.

[0021] The amino acid sequence $(X_1)_n X_2 X_3 X_4 X_5 G X_6 P (X_7)_m$ is considered to be a mimotope of the epitope of alpha-synuclein comprising the amino acid sequence KNEEGAP. According to the present invention the term "mimotope" refers to a molecule which has a conformation that has a topology equivalent to the epitope of which it is a mimic. The mimotope binds to the same antigen-binding region of an antibody which binds immunospecifically to a desired antigen. The mimotope will elicit an immunological response in a host that is reactive to the antigen to which it is a mimic. The mimotope may also act as a competitor for the epitope of which it is a mimic in *in vitro* inhibition assays (e.g. ELISA inhibition assays) which involve the epitope and an antibody binding to said epitope. However, a mimotope of the present invention may not necessarily prevent or compete with the binding of the epitope of which it is a mimic in an *in vitro* inhibition assay although it is capable to induce a specific immune response when administered to a mammal.

[0022] As used herein, the term "epitope" refers to an immunogenic region of an antigen which is recognized by a particular antibody molecule. In general, an antigen will possess one or more epitopes, each capable of binding an antibody that recognizes the particular epitope.

[0023] The mimotopes of the present invention can be synthetically produced by chemical synthesis methods which are well known in the art, either as an isolated peptide or as a part of another peptide or polypeptide. Alternatively, the peptide mimotope can be produced in a microorganism which produces the peptide mimotope which is then isolated and if desired, further purified. The peptide mimotope can be produced in microorganisms such as bacteria, yeast or fungi, in eukaryote cells such as a mammalian or an insect cell, or in a recombinant virus vector such as adenovirus, poxvirus, herpesvirus, Simliki forest virus, baculovirus, bacteriophage, sindbis virus or sendai virus. Suitable bacteria for producing the peptide mimotope include E.coli, B.subtilis or any other bacterium that is capable of expressing peptides such as the peptide mimotope. Suitable yeast types for expressing the peptide mimotope include Saccharomyces cerevisiae, Schizosaccharomyces pombe, Candida, Pichia pastoris or any other yeast capable of expressing peptides. Corresponding methods are well known in the art. Also methods for isolating and purifying recombinantly produced peptides are well known in the art and include e.g. gel filtration, affinity chromatography, ion exchange chromatography etc.

[0024] To facilitate isolation of the peptide mimotope, a fusion polypeptide may be made wherein the peptide mimotope is translationally fused (covalently linked) to a heterologous polypeptide which enables isolation by affinity chromatography. Typical heterologous polypeptides are His-Tag (e.g. His₆; 6 histidine residues), GST-Tag (Glutathione-S-transferase) etc.. The fusion polypeptide facilitates not only the purification of the mimotopes but can also prevent the mimotope polypeptide from being degraded during purification. If it is desired to remove the heterologous polypeptide after purification the fusion polypeptide may comprise a cleavage site at the junction between the peptide mimotope and the heterologous polypeptide. The cleavage site consists of an amino acid sequence that is cleaved with an enzyme specific for the amino acid sequence at the site (e.g. proteases).

[0025] According to a preferred embodiment of the present invention, X_2 is an amino acid residue selected from the group consisting of lysine (K) and arginine (R) and/or X_6 is alanine (A).

[0026] According to a particularly preferred embodiment of the present invention, the peptide or polypeptide comprises an amino acid sequence selected from the group consisting of $(X_1)_n$ KNDEGAP $(X_7)_m$, $(X_1)_n$ ANEEGAP $(X_7)_m$, $(X_1)_n$ KAEAGAP $(X_7)_m$, $(X_1)_n$ KNAEGAP $(X_7)_m$, $(X_1)_n$ RNEEGAP $(X_7)_m$, $(X_1)_n$ HNEEGAP $(X_7)_m$, $(X_1)_n$ KNEDGAP $(X_7)_m$, $(X_1)_n$ KQEEGAP $(X_7)_m$, $(X_1)_n$ KSEEGAP $(X_7)_m$, $(X_1)_n$ KNDDGAP $(X_7)_m$, $(X_1)_n$ RNDEGAP $(X_7)_m$, $(X_1)_n$ RNEDGAP $(X_7)_m$, $(X_1)_n$ RQEEGAP $(X_7)_m$, $(X_1)_n$ RSEEGAP $(X_7)_m$, $(X_1)_n$ ANEDGAP $(X_7)_m$, $(X_1)_n$ ANEDGAP $(X_7)_m$, $(X_1)_n$ HSEEGAP $(X_7)_m$, $(X_1)_n$ ASEEGAP $(X_7)_m$, $(X_1)_n$ HNEDGAP $(X_7)_m$, $(X_1)_n$ HNEDGAP $(X_7)_m$, $(X_1)_n$ RNAEGAP $(X_7)_m$, $(X_1)_n$ HNAEGAP $(X_7)_m$, $(X_1)_n$ KSAEGAP $(X_7)_m$, $(X_1)_n$ KSDEGAP $(X_7)_m$, $(X_1)_n$ KSEDGAP $(X_7)_m$, $(X_1)_n$ RQDEGAP $(X_7)_m$, $(X_1)_n$ RQEDGAP $(X_7)_m$, $(X_1)_n$ HSAEGAP $(X_7)_m$, $(X_1)_n$ RSAEGAP $(X_7)_m$, $(X_1)_n$ RSDEGAP $(X_7)_m$, $(X_1)_n$ RSEDGAP $(X_7)_m$, $(X_1)_n$ HSDEGAP $(X_7)_m$, $(X_1)_n$ HSEDGAP $(X_7)_m$, $(X_1)_n$ RQDDGAP $(X_7)_m$, preferably $(X_1)_n$ KNDEGAP $(X_2)_m$, $(X_1)_n$ RNEEGAP $(X_2)_m$, $(X_1)_n$ KNDEGAP $(X_2)_m$, $(X_1)_n$ KNAEGAP $(X_2)_m$, $(X_1)_n$ KSDEGAP $(X_2)_m$, $(X_1)_n$ RNAEGAP $(X_2)_m$ Or $(X_1)_n$ RSEEGAP $(X_2)_m$.

[0027] The peptides and polypeptides of the present invention may also be modified at or nearby their N- and/or C-termini so that at said positions a cysteine residue is bound thereto. In a preferred embodiment terminally positioned (located at the N- and C-termini of the peptide) cysteine residues are used to crosslink said molecules with carrier molecules such as KLH or to cyclize the peptides through a disulfide bond. Therefore, n and/or m are preferably 1 and X₁ and/or X₇ are preferably cysteine (C).

[0028] The mimotopes of the present invention may also be used in various assays and kits, in particular in immunological assays and kits. Therefore, it is particularly preferred that the peptides and polypeptides of the present invention may be part of another peptide or polypeptide, particularly an enzyme which is used as a reporter in immunological assays. Such reporter enzymes include e.g. alkaline phosphatase or horseradish peroxidase.

[0029] The alpha-synuclein mimotopes according to the present invention preferably are antigenic polypeptides which in their amino acid sequence vary from the amino acid sequence of alpha-synuclein or of fragments of alpha-synuclein. In this respect, the inventive mimotopes may not only comprise amino acid substitutions of one or more naturally occurring amino acid residues but also of one or more non-natural amino acids (i.e. not from the 20 "classical" amino acids) or they may be completely assembled of such non-natural amino acids. Moreover, the inventive antigens which induce anti-alpha-synuclein antibodies may be assembled of D- or L- amino acids or of combinations of DL- amino acids and, optionally, they may have been changed by further modifications, ring closures or derivatizations. Suitable anti-alpha-synuclein-antibody-inducing antigens may be provided from commercially available peptide libraries. Preferably, these peptides are at least 7 amino acids, and preferred lengths may be up to 16, preferably up to 14 or 20 amino acids residues (e.g. 7 or 8 to 20, 7 or 8 to 16 etc.). Thus, the peptide or polypeptide of the present invention comprises 7 to 30, preferably 7 to 20, more preferably 7 to 16, most preferably 8, amino acid residues. According to the invention, however, also longer peptides may very well be employed as anti-alpha-synuclein-antibody-inducing antigens. Furthermore the mimotopes of the present invention may also be part of a polypeptide and consequently comprising at their N- and/or C-terminus at least one further amino acid residue.

[0030] For preparing alpha-synuclein mimotopes (i.e. anti-alpha-synuclein-antibody-inducing antigens), of course also phage libraries, peptide libraries are suitable, for instance produced by means of combinatorial chemistry or obtained by means of high throughput screening techniques for the most varying structures (Display: A Laboratory Manual by Carlos F. Barbas (Editor), et al.; Willats WG Phage display: practicalities and prospects. Plant Mol. Biol. 2002 Dec.; 50(6):837-54).

[0031] Furthermore, according to the invention also anti-alpha-synuclein-antibody-inducing antigens based on nucleic acids ("aptamers") may be employed, and these, too, may be found with the most varying (oligonucleotide) libraries (e.g. with 2-180 nucleic acid residues) (e.g. Burgstaller et al., Curr. Opin. Drug Discov. Dev. 5(5) (2002), 690-700; Famulok et al., Acc. Chem. Res. 33 (2000), 591-599; Mayer et al., PNAS 98 (2001), 4961-4965, etc.). In anti-alpha-synuclein-antibody-inducing antigens based on nucleic acids, the nucleic acid backbone can be provided e.g. by the natural phosphor-diester compounds, or also by phosphorotioates or combinations or chemical variations (e.g. as PNA), wherein as bases, according to the invention primarily U, T, A, C, G, H and mC can be employed. The 2'-residues of the nucleotides which can be used according to the present invention preferably are H, OH, F, Cl, NH₂, O-methyl, O-ethyl, O-propyl or O-butyl, wherein the nucleic acids may also be differently modified, i.e. for instance with protective groups, as they are commonly employed in oligonucleotide synthesis. Thus, aptamer-based anti-alpha-synuclein-antibody-inducing antigens are also preferred anti-alpha-synuclein-antibody-inducing antigens within the scope of the present invention.

[0032] According to the present invention the term "synucleinopathy" includes all neurodegenerative disorders characterized by pathological synuclein aggregations. Several neurodegenerative disorders including Parkinson's Disease (PD), Lewy Body Disease (LBD), Diffuse Lewy Body Disease (DLBD), Dementia with Lewy Bodies (DLB), Parkinsonism with Dementia (PDD), Multiple System Atrophy (MSA) and Neurodegeneration with Brain Iron Accumulation type I (NBIA Type I) are collectively grouped as synucleinopathies.

[0033] The peptides and polypeptides according to the present invention may be employed not only for treating synucleinopathies but also to prevent said diseases in individuals being at risk of developing a synucleinopathy (e.g. predisposed, for example genetically predisposed, to developing a synucleinopathy).

[0034] The abbreviations for the amino acid residues disclosed in the present invention follow the IUPAC recommendations:

Amino Acid	3-Letter Code	1-Letter Code
Alanine	Ala	A
Arginine	Arg	R
Asparagine	Asn	N
Aspartic	Asp	D

(continued)

Amino Acid	3-Letter Code	1-Letter Code
Cysteine	Cys	C
Glutamic	Glu	E
Glutamine	Gln	Q
Glycine	Gly	G
Histidine	His	H
Isoleucine	Ile	I
Leucine	Leu	L
Lysine	Lys	K
Methionine	Met	M
Phenylalanine	Phe	F
Proline	Pro	P
Serine	Ser	S
Threonine	Thr	T
Tryptophan	Trp	W
Tyrosine	Tyr	Y
Valine	Val	V

[0035] The peptides and polypeptides of the present invention may also be part of a polypeptide comprising 7 to 30 amino acid residues. Consequently n and m may independently be an integer selected from the group of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 20 and 25.

[0036] The at least one peptide or polypeptide according to the present invention may consist of the amino acid sequence $(X_1)_n X_2 X_3 X_4 X_5 G X_6 P (X_7)_m$, wherein n and m are independently 0 or 1 or being part of a polypeptide which comprises at least 7 amino acid residues, preferably at least 10 amino acid residues, more preferably at least 15 amino acid residue, and/or a maximum of 50 amino acid residues, preferably a maximum of 30 amino acid residues, more preferably of 16 amino acid residues.

[0037] Surprisingly, it turned out that the compounds according to the present invention comprising or consisting of the amino acid sequences listed above are particularly suited to be used for the manufacture of a medicament to be used to treat or prevent synucleinopathies. These peptides (mimotopes) are able to induce the in vivo formation of antibodies directed to the original epitope of human alpha-synuclein comprising the amino acid sequence KNEEGAP and human alpha-synuclein protein itself. Said peptides (mimotopes) are, however, not able to induce immune reactivity against human beta-synuclein protein. The peptide induced antibodies are responsible for the removal of alpha-synuclein (which is involved in the formation of alpha-synuclein aggregates, Lewy bodies) and/or for the dissolution of alpha-synuclein aggregates (Lewy bodies) in an individual.

[0038] The peptides and polypeptides according to the present invention may be used for the preparation of a medicament, in particular a vaccine, which can be used to treat alpha-synucleinopathy, whereby the medicament is particularly suited to treat synucleinopathy selected from the group consisting of Parkinson's Disease (PD), Lewy Body Disease (LBD), Diffuse Lewy Body Disease (DLBD), Dementia with Lewy Bodies (DLB), Parkinson-ism with Dementia (PDD), Multiple System Atrophy (MSA) and Neurodegeneration with Brain Iron Accumulation type I (NBIA Type I).

[0039] According to a preferred embodiment of the present invention the at least one peptide or polypeptide is coupled to a pharmaceutically acceptable carrier, preferably KLH (Keyhole Limpet Hemocyanin), tetanus toxoid, albumin-binding protein, bovine serum albumin, a dendrimer (MAP; Biol. Chem. 358: 581), peptide linkers (or flanking regions) as well as the adjuvant substances described in Singh et al., Nat. Biotech. 17 (1999), 1075-1081 (in particular those in Table 1 of that document), and O'Hagan et al., Nature Reviews, Drug Discovery 2 (9) (2003), 727-735 (in particular the endogenous immuno-potentiating compounds and delivery systems described therein), and others or mixtures thereof. The conjugation chemistry (e.g. via heterobifunctional compounds such as GMBS and of course also others as described in "Bioconjugate Techniques", Greg T. Hermanson) in this context can be selected from reactions known to the skilled man in the art. Moreover, the vaccine composition may be formulated with an adjuvant, preferably a low soluble aluminium

composition, in particular aluminium hydroxide. Of course, also adjuvants like MF59 aluminium phosphate, calcium phosphate, cytokines (e.g., IL-2, IL-12, GM-CSF), saponins (e.g., QS21), MDP derivatives, CpG oligos, IC31, LPS, MPL, polyphosphazenes, emulsions (e.g., Freund's, SAF), liposomes, virosomes, iscoms, cochleates, PLG microparticles, poloxamer particles, virus-like particles, heatlabile enterotoxin (LT), cholera toxin (CT), mutant toxins (e.g., LTK63 and LTR72), microparticles and/or polymerized liposomes may be used.

[0040] The peptide or polypeptide of the present invention is preferably bound to the carrier or adjuvant via a linker, which is selected from the group consisting of NHS-poly (ethylene oxide) (PEO) (e.g. NHS-PEO₄-maleimide).

[0041] A vaccine which comprises the present compound (mimotope) and the pharmaceutically acceptable carrier may be administered by any suitable mode of application, e.g. i.d., i.p., i.m., intranasally, orally, subcutaneously, etc. and in any suitable delivery device (O'Hagan et al., Nature Reviews, Drug Discovery 2 (9), (2003), 727-735). The compound of the present invention is preferably formulated for subcutaneous, intradermal or intramuscular administration (see e.g. "Handbook of Pharmaceutical Manufacturing Formulations", Sarfaraz Niazi, CRC Press Inc, 2004).

[0042] Typically, the vaccine contains the compound according to the invention in an amount of from 0.1 ng to 10 mg, preferably 10 ng to 1 mg, in particular 100 ng to 100 µg, or, alternatively, e.g. 100 fmol to 10 µmol, preferably 10 pmol to 1 µmol, in particular 100 pmol to 100 nmol. Typically, the vaccine may also contain auxiliary substances, e.g. buffers, stabilizers etc.. Preferably, such auxiliary substances, e.g. a pharmaceutically acceptable excipient, such as water, buffer and/or stabilisers, are contained in an amount of 1 to 99 % (weight), more preferred 5 to 80% (weight), especially 10 to 70 % (weight). Possible administration regimes include a weekly, biweekly, fourweekly (monthly) or bimonthly treatment for about 1 to 12 months; however, also 2 to 5, especially 3 to 4, initial vaccine administrations (in one or two months), followed by booster vaccinations 6 to 12 months thereafter or even years thereafter are preferred - besides other regimes already suggested for other vaccines.

[0043] Another aspect of the present invention relates to a peptide having an amino acid sequence selected from the group consisting of (X₁)_nKNDEGAP(X₇)_m, (X₁)_nANEEGAP(X₇)_m, (X₁)_nKAEEGAP(X₇)_m, (X₁)_nKNAEGAP(X₇)_m, (X₁)_nRNEEGAP(X₇)_m, (X₁)_nHNEEGAP(X₇)_m, (X₁)_nKNEDGAP(X₇)_m, (X₁)_nKQEEGAP(X₇)_m, (X₁)_nKSEEGAP(X₇)_m, (X₁)_nKND-DGAP(X₇)_m, (X₁)_nRNDEGAP(X₇)_m, (X₁)_nRNEDGAP(X₇)_m, (X₁)_nRQEEGAP(X₇)_m, (X₁)_nRSEEGAP(X₇)_m, (X₁)_nANDEGAP(X₇)_m, (X₁)_nANEDGAP(X₇)_m, (X₁)_nHSEEGAP(X₇)_m, (X₁)_nASEEGAP(X₇)_m, (X₁)_nHNEDGAP(X₇)_m, (X₁)_nHNDEGAP(X₇)_m, (X₁)_nRNAEGAP(X₇)_m, (X₁)_nHNAEGAP(X₇)_m, (X₁)_nKSAEGAP(X₇)_m, (X₁)_nKSDEGAP(X₇)_m, (X₁)_nKSEDGAP(X₇)_m, (X₁)_nRQDEGAP(X₇)_m, (X₁)_nRQEDGAP(X₇)_m, (X₁)_nHSAEGAP(X₇)_m, (X₁)_nRSAEGAP(X₇)_m, (X₁)_nRSDEGAP(X₇)_m, (X₁)_nRSEDGAP(X₇)_m, (X₁)_nHSDEGAP(X₇)_m, (X₁)_nHSEDGAP(X₇)_m and (X₁)_nRQDDGAP(X₇)_m, wherein X₁ and X₇ is cysteine and n and m, independently, are 0 or 1.

[0044] According to a preferred embodiment of the present invention the peptide is coupled to a pharmaceutically acceptable carrier, preferably KLH (Keyhole Limpet Hemocyanin).

[0045] The pharmaceutical formulation according to the present invention, which can be formulated as a vaccine for, e.g., subcutaneous, intradermal and/or intramuscular administration, may be used in the treatment of any kind of synucleinopathy.

[0046] The present invention is further illustrated in the following figures and examples, however, without being restricted thereto.

[0047] Fig. 1 shows the sequence of full length alpha-synuclein (140aa; swiss prot entry: P37840) and the sequence used to create a monoclonal antibody for detection of full length alpha-synuclein as well as C-terminally truncated and modified versions thereof. The peptide at position 100-109 used for generation of the monoclonal antibody is underlined. The peptide (p4453) was coupled to a C at C-terminal position.

[0048] Fig. 2 shows detection of alpha-synuclein by ELISA using the generated monoclonal antibody specific for human alpha-synuclein at position 100-109. The monoclonal antibody 12-9-9 was generated and tested for its specificity to synucleins in ELISA. Alpha-synuclein (p4446) and p4453 the human epitope are detected. The negative control protein p4447 (beta-synuclein) is not detected.

[0049] Fig. 3 shows the definition of the minimal epitope of the monoclonal antibody 12-9-9 by ELISA.

[0050] The peptides p4446 (alpha-synuclein), p4453 (human epitope used to create the antibody tested) are detected by the anti-body. The original epitope p4453 was N- or C-terminally truncated and used for ELISA to define the minimal epitope required for specific binding. The peptides p5399 and p5403 lost binding to the monoclonal antibody 12-9-9. Thus the minimal sequence needed for binding of 12-9-9 is predicted as KNEEGAP located at position 102-108 of alpha-synuclein, while truncation of one of the flanking amino acids abolished the binding. Data are presented in a linear scale.

[0051] Fig. 4 shows detection of epitopes and mimotopes by ELISA using a monoclonal specific for human alpha-synuclein at position 100-109. Alpha-Synuclein as well as the peptides p5436 (human minimal epitope) and the mimotopes p5439 are detected similarly by the monoclonal antibody 12-9-9. The mimotope p5440 is not detected, while mimotope p5444 is detected much weaker than the human epitope by the monoclonal antibody 12-9-9.

[0052] Fig. 5 shows the induction of immune response against alpha-synuclein after peptide immunization. Sera of immunized mice (p5436 to p5590) show titers against alpha-synuclein after 3 vaccinations. Sera of immunized mice (p5463 to p5466) do not detect alpha synuclein (Titers measured in ELISA are around or below 1:50 half-max). Class

of immunogenicity was defined as follows: Class 2: peptides inducing an immune response with OD halfmax higher than 1:1000. Class 1: peptides inducing an immune response with OD halfmax between 1000 and 51. Class 0: peptides inducing no or an very low immune response with OD halfmax around 50 or lower.

EXAMPLES:

[0053] To identify peptides and polypeptides which can be used to treat and/or prevent synucleopathies an antibody was used, which is able to detect the human alpha-synuclein-derived amino acid sequence LGKNEEGAPQ (= original epitope, SEQ ID No. 3, p4453) and full length human alpha-synuclein (SEQ ID No. 1, p4446). It does not recognize human beta-synuclein (SEQ ID No. 2, p4447; accession number Q16143: mdvfmkgism akegvvaae ktkqgvtea ektkegv-lyv gsktregvvq gvasvaektk eqashlggav fsgagniaaa tglvkreefp tdlkpeeva eaaeepliep lmepegesy e dppqeeyqey epea). The antibody may be a monoclonal or polyclonal antibody preparation or any antibody part or derivative thereof and binds specifically to the LGKNEEGAPQ epitope of human alpha-synuclein, i. e. it does bind to peptide and full length protein but does not bind to human beta-synuclein. The mimotopes are identified and further characterised with such monoclonal antibodies (detecting a sequence within amino acids 100-109 of the human alpha-synuclein protein) and peptide libraries.

Example 1: Generation of monoclonal antibodies to specifically detect original human alpha-synuclein epitope LG-KNEEGAPQC SEQ ID No. 3, p4453 and human alpha-synuclein but not human beta-synuclein.

[0054] A monoclonal antibody derived from the fusion "AFFiRiS 6": Balb/c mice (Charles River) were immunized with original alpha-synuclein epitope LGKNEEGAPQ-C coupled to BTG (bovine thyroglobuline) and CFA (complete Freund's adjuvant; first injection) as well as IFA (incomplete Freund's adjuvant; 3 booster injections) as adjuvant. LGKNEEGAPQ-peptide-specific, antibody-producing hybridomas are detected by ELISA (LGKNEEGAPQ-peptide-coated ELISA plates). Human alpha-synuclein (recombinant protein, p4446) is used as positive control peptide: hybridomas recognizing the recombinant protein immobilized on ELISA plates are included because they are binding both peptide and full length alpha-synuclein specifically. Human beta-synuclein (recombinant protein, p4447) is used as negative control peptide: hybridomas recognizing both recombinant proteins immobilized on ELISA plates are excluded because they do not distinguish between the two different synuclein proteins. The Hybridoma clone (12-9-9; IgG1, kappa) was analysed for specific detection of the natural human alpha-synuclein epitope LGKNEEGAPQ. 12-9-9 recognizes the injected epitope as well as full length alpha-synuclein protein (recombinant protein; obtained from rPeptide, Bogart, GA, USA) in ELISA (see Fig. 2). It however does not detect beta-synuclein protein (recombinant protein, obtained from rPeptide, Bogart, GA, USA) in ELISA (see Fig. 2). Subsequently, the minimal epitope required for binding of the antibody has been defined by ELISA using the peptides p4446, p4453, p5397, p5398, p5399, p5400, p5401, p5402, p5403, p5404, p5405, p5406 (see Fig. 3) and p5436 (see Fig. 4). p4446, p4453, p5397, p5398 and p5402 as well as p5436 retained full binding capacities whereas p5399, p5400, p5401, p5403, p5404, p5405 and p5406 lost binding to 12-9-9. Thus the minimal required epitope for binding has been defined as KNEEGAP.

Example 2: Phage Display, in vitro binding and inhibition ELISA

[0055] Phage Display libraries used in this example were: Ph.D. 7: New England BioLabs E8102L (linear 7mer library), Ph.D. 12: New England BioLabs E8111L (linear 12mer library) and Ph.D. C7C: New England BioLabs E8120L (a disulfide-constrained heptapeptide library) Phage Display was done according to manufacturer's protocol (www.neb.com). After 2 or 3 subsequent rounds of panning, single phage clones were picked and phage supernatants were subjected to ELISA on plates coated with the antibody that was used for the panning procedure. Phage clones that were positive in this ELISA (strong signal for the target, but no signal for unspecific control) were sequenced. From DNA sequences, peptide sequences were deduced. These peptides were synthesized and characterised in binding and inhibition ELISA. To some peptides additional AA were attached to the C-terminus. Additionally, some novel mimotopes were created by combining sequence information from mimotopes identified in the screen. Both groups containing newly designed mimotopes were used to support the identification of a consensus sequence for a mimotope vaccination.

1. In vitro binding assay (ELISA)

[0056] Peptides derived from Phage Display as well as N-terminally truncated variants thereof were coupled to BSA and bound to ELISA plates (1µM) and subsequently incubated with the monoclonal antibody that was used for the screening procedure to analyse binding capacity of identified peptides (see Fig. 4).

2. *In vitro* inhibition assay (ELISA)

[0057] Different amounts of peptides (concentrations ranging from 400 µg/ml to 3 µg/ml (serial dilutions), derived from Phage Display were incubated with the monoclonal antibody that was used for the screening procedure. Peptides diminishing subsequent binding of the antibody to the original human alpha-synuclein epitope (p5436) and the human alpha-synuclein protein (p4446) coated on ELISA plates were considered as inhibiting in this assay.

Example 3: in vivo testing of mimotopes: analysis of immunogenicity

1. In vivo testing of mimotopes

[0058] Inhibiting as well as non-inhibiting peptides were coupled to KLH and injected into mice (wildtype C57/B16 or BalbC mice; subcutaneous injection into the flank) together with an appropriate adjuvant (aluminium hydroxide). Animals were vaccinated 3 times in biweekly intervals and sera were taken biweekly as well. Titers to injected peptides as well as to an irrelevant peptide were determined with every serum. Titers against the recombinant human alpha-synuclein protein and recombinant human beta-synuclein were determined starting with Serum 2 respectively. In general sera were analysed by reaction against peptides coupled to Bovine Serum Albumin (BSA) and recombinant full length proteins which were immobilised on ELISA plates. Titers were determined using anti mouse IgG specific antibodies. For examples of immunogenicity against injected peptide and alpha-synuclein see Table 5 and Table 6.

2. Results

[0059]

2.1. Identification of an alpha-synuclein specific mAB: Figure 2 depicts the characterisation of the alpha-synuclein specific monoclonal antibody 12-9-9 (IgG1, kappa) derived from fusion Affiris 6.

2.2. Screening for alpha-synuclein specific Mimotopes:

2.2.1. Phage Display PhD 7, PhD12 and PhD C7C and mutational Screen

2.2.1.1. Screening with monoclonal antibody directed against LGKNEEGAPQ

By screening of PhD 7, PhD12 and PhD C7C phage display libraries and selective displacement of single amino acids a total of 60 sequences was identified (see Table 1; ID18-77).

Table 1 shows examples all peptides used.

SEQ ID	peptide No	sequence
1	p4446	Alpha-synuclein (Fig. 1; P37840)
2	p4947	Beta-Synuclein; Q16143
3	p4453	LGKNEEGAPQC
4	p4454	MGKGEEGYPQC
5	p5397	GKNEEGAPQC
6	p5398	KNEEGAPQC
7	p5399	NEEGAPQC
8	p5400	EEGAPQC
9	p5401	EGAPQC
10	p5402	LGKNEEGAPC
11	p5403	LGKNEEGAC
12	p5404	LGKNEEGC
13	p5405	LGKNEEC
14	p5406	LGKNEC

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SEQ ID	peptide No	sequence
15	p5435	CKNEEGAP
16	p5436	KNEEGAPC
17	p1253	DAEFRHDSGY-C
18	p5437	ANEEGAPC
19	p5438	KAEEGAPC
20	p5439	KNAEGAPC
21	p5440	KNEAGAPC
22	p5441	KNEEAAPC
23	p5442	KNEEGAAC
24	p5443	RNEEGAPC
25	p5444	HNEEGAPC
26	p5445	KNDEGAPC
27	p5446	KNEDGAPC
28	p5447	KQEEGAPC
29	p5448	KSEEGAPC
30	p5449	KNDDGAPC
31	p5461	KPSFKNEC
32	p5462	QPSFAMEC
33	p5463	SPSFKQEC
34	p5464	TPSWKGEC
35	p5465	DPSFALEC
36	p5466	LPSFRLEC
37	p5467	EPNSRMDC
38	p5468	QPSSKLDC
39	p5469	HIHQSKFFDAPPC
40	p5547	QASFAMEC
41	p5548	TASWKGEC
42	p5549	QASSKLDC
43	p5550	QPAFAMEC
44	p5551	TPAWKGEC
45	p5552	QPASKLDC
46	p5553	QPSFAMAC
47	p5554	TPSWKGAC
48	p5555	QPSSKLAC
49	p5556	APSWKGEC
50	p5557	TPSAKGEC
51	p5558	TPSWAGEC
52	p5559.	TPSWKAEC

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SEQ ID	peptide No	sequence
53	p5560	CTPSWKGE
54	p5587	RNDEGAPC
55	p5588	RNEDGAPC
56	p5589	RQEEGAPC
57	p5590	RSEEGAPC
58	p5591	ANDEGAPC
59	p5592	ANEDGAPC
60	p5593	HSEEGAPC
61	p5594	ASEEGAPC
62	p5595	HNEDGAPC
63	p5596	HNDEGAPC
64	p5597	RNAEGAPC
65	p5598	HNAEGAPC
66	p5599	KSAEGAPC
67	p5600	KSDEGAPC
68	p5601	KSEDGAPC
69	p5602	RQDEGAPC
70	p5603	RQEDGAPC
71	p5604	HSAEGAPC
72	p5605	RSAEGAPC
73	p5606	RSDEGAPC
74	p5607	RSEDGAPC
75	p5608	HSDEGAPC
76	p5609	HSEDGAPC
77	p5610	RQDDGAPC

Table 2 shows examples of peptides and their binding capacity as compared to the original epitope.

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internal number	Sequence	Binding	remark
p5435	CKNEEGAP	2	original
p5436	KNEEGAPC	2	original
p5437	ANEEGAPC	2	mimotope
p5438	KAEEGAPC	1	mimotope
p5439	KNAEGAPC	2	mimotope
p5443	RNEEGAPC	2	mimotope
p5444	HNEEGAPC	2	mimotope
p5445	KNDEGAPC	2	mimotope
p5446	KNEDGAPC	2	mimotope

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(continued)

	internal number	Sequence	Binding	remark
5	p5447	KQEEGAPC	2	mimotope
	p5448	KSEEGAPC	2	mimotope
	p5449	KNDDGAPC	2	mimotope
	p5398	KNEEGAPQC	2	original
10	p5402	LGKNEEGAPC	2	original
	p5397	GKNEEGAPQC	2	original
	p4454	MGKGEEGYPC	2	original mouse
15	p4453	LGKNEEGAPQC	2	original
	p5461	KPSFKNEC	2	mimotope
	p5462	QPSFAMEC	2	mimotope
	p5463	SPSFKQEC	2	mimotope
20	p5464	TPSWKGEC	2	mimotope
	p5465	DPSFALEC	2	mimotope
	p5466	LPSFRLEC	1	mimotope
25	p5468	QPSSKLDC	2	mimotope
	p5547	QASFAMEC	1	mimotope
	p5548	TASWKGEC	2	mimotope
	p5549	QASSKLDC	1	mimotope
30	p5551	TPAWKGEC	1	mimotope
	p5556	APSWKGEC	2	mimotope
	p5558	TPSWAGEC	2	mimotope
35	p5560	CTPSWKGE	2	mimotope
	p5587	--RNDEGAPC	2	mimotope
	p5588	--RNEDGAPC	2	mimotope
	p5589	--RQEEGAPC	2	mimotope
40	p5590	--RSEEGAPC	2	mimotope
	p5591	--ANDEGAPC	2	mimotope
	p5593	--HSEEGAPC	1	mimotope
45	p5594	--KSEEGAPC	1	mimotope
	p5596	--HNDEGAPC	2	mimotope
	p5597	--RNAEGAPC	2	mimotope
	p5598	--HNAEGAPC	1	mimotope
50	p5599	--KSAEGAPC	2	mimotope
	p5600	--KSDEGAPC	2	mimotope
	p5601	--KSEDGAPC	2	mimotope
55	p5602	--RQDEGAPC	2	mimotope
	p5603	--RQEDGAPC	2	mimotope
	p5604	--HSAEGAPC	1	mimotope

(continued)

internal number	Sequence	Binding	remark
p5605	--RSAEGAPC	2	mimotope
p5606	--RSDEGAPC	2	mimotope
p5607	--RSEDGAPC	2	mimotope
p5608	--HSDEGAPC	1	mimotope
p5610	--RQDDGAPC	2	mimotope
p4446		2	α -Syn
Table 2: Examples of alpha-synuclein epi- and mimotopes binding to the monoclonal Antibody 12-9-9 the binding capacity is coded by the following code: 0: no binding to 12-9-9 detectable in ELISA 1: weak binding: binding of mimotope weaker compared to the minimal original sequence p5436 2: strong binding: binding of mimotope similar to the minimal original sequence p5436			

2.3. In vitro characterisation of mimotopes identified in a screen (Phage Display and peptide screening) with a monoclonal antibody directed against alpha-synuclein:

Fig. 2 and 3 show representative examples for binding and inhibition assays used to characterise mimotopes in vitro. Data obtained are summarised in Tables 2 and 3 respectively.

Table 3: Inhibition assay

designation	Sequence	Competition	remark
p5435	CKNEEGAP	1	original
p5436	KNEEGAPC	2	original
p5439	KNAEGAPC	1	mimotope
p5443	RNEEGAPC	2	mimotope
p5445	KNDEGAPC	2	mimotope
p5446	KNEDGAPC	1	mimotope
p5448	KSEEGAPC	1	mimotope
p5449	KNDDGAPC	1	mimotope
p5398	KNEEGAPQC	2	original
p5402	LGKNEEGAPC	2	original
p5397	GKNEEGAPQC	2	original mouse
p5464	TPSWKGEC	1	mimotope
p5548	TASWKGEC	1	mimotope
p5556	APSWKGEC	1	mimotope
p5557	TPSAKGEC	1	mimotope
p5587	RNDEGAPC	2	mimotope
p5588	RNEDGAPC	1	mimotope
p5590	RSEEGAPC	1	mimotope
p5597	RNAEGAPC	1	mimotope
p5600	KSDEGAPC	1	mimotope
p5602	RQDEGAPC	1	mimotope

(continued)

designation	Sequence	Competition	remark
p5603	RQEDGAPC	1	mimotope
Table 3: Alpha-synuclein mimotopes identified in this invention giving positive results in inhibition assays Legend to Table 3: the competition capacity is coded by the following code: 0: no competition of 12-9-9 detectable in ELISA 1: weak competition: competition of mimotope weaker compared to the minimal original sequence p5436 2: strong competition: competition of mimotope similar to the minimal original sequence p5436			

Table 4: Non-Mimotope peptides and proteins:

SEQ ID No.	designation	sequence
1	p4446	Alpha-synuclein
2	p4447	Beta-Synuclein
3	p4453	LGKNEEGAPQC
4	p4454	MGKGEEGYPQC
5	p5397	GKNEEGAPQC
6	p5398	KNEEGAPQC
7	p5399	NEEGAPQC
8	p5400	EEGAPQC
9	p5401	EGAPQC
10	p5402	LGKNEEGAPC
11	p5403	LGKNEEGAC
12	p5404	LGKNEEGC
13	p5405	LGKNEEC
14	p5406	LGKNEC
15	p5435	CKNEEGAP
16	p5436	KNEEGAPC
17	p1253	DAEFRHDSGY-C

2.4. *In vivo* characterisation of mimotopes identified in screening Phage Display Libraries with a monoclonal antibody directed against alpha-synuclein:

Female C57/B16 mice or BalbC, 5-6 mice per group, were subcutaneously immunized with 30 µg peptide coupled to KLH. Control groups were injected with PBS or the original epitope. As adjuvant alum was used. The peptides administered were all able to bind to monoclonal antibodies specifically binding aa100-109 of human alpha-synuclein although some of the peptides did inhibit the binding of the original epitope to its parental antibody in vitro only weakly (in an in vitro inhibition assay). The in vitro ELISA assay to determine the antibody titer was performed with sera of single mice (see Tab. 5) after each vaccination in a two week interval. The wells of the ELISA plate were coated with mimotope-BSA conjugates. The positive control was performed by reaction of the parental antibody with the respective mimotope-BSA conjugate. The detection was performed with anti-mouse IgG. Additionally, recombinant proteins were immobilised on ELISA plates and sera reacted accordingly. For all mimotopes tested in C57/B16 mice or BalbC, antibodies reacting to the individual injected peptide could be detected after repeated vaccination. Although not all mice induced higher titer against alpha-synuclein (see tab. 5 for examples).

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Table 5: Induction of immune response is indicated by the titer against injected peptide (p4446). Titer was measured by ELISA and indicated as OD halfmax.

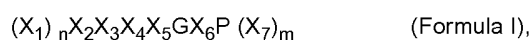
designation	Sequence	Titer in animals OD halfmax	remark
		injected pep.	
p5436	KNEEGAPC	10000	original
p5439	KNAEGAPC	8000	mimotope
p5443	RNEEGAPC	9000	mimotope
p5445	KNDEGAPC	26000	mimotope
p5402	LGKNEEGAPC	20000	original
p4454	MGKGEEGYPQC	17000	original mouse

Table 6: class of immunogenicity of mimotopes against a Syn

designation	sequence	class of immunogenicity: a Syn	remark
p5402	LGKNEEGAP-C	2	original
p5436	KNEEGAPC	2	original
p5445	KNDEGAPC	2	mimotope
p5443	RNEEGAPC	1	mimotope
p5587	RNDEGAPC	1	mimotope
p5439	KNAEGAPC	1	mimotope
p5600	KSDEGAPC	1	mimotope
p4454	MGKGEEGYPQC	1	original mouse
p5597	RNAEGAPC	1	mimotope
p5590	RSEEGAPC	1	mimotope
p5463	SPSFKQEC	0	mimotope
p5556	APSWKGEC	0	mimotope
p5462	QPSFAMEC	0	mimotope
p5464	TPSWKGEC	0	mimotope
p5468	QPSSKLDC	0	mimotope
p5461	KPSFKNEC	0	mimotope
p5465	DPSFALEC	0	mimotope
p5466	LPSFRLEC	0	mimotope
Class of immunogenicity: peptides were ranked according to their capacity to induce an immune response 2: peptides inducing an immune response with OD halfmax higher than 1000. 1: peptides inducing an immune response with OD halfmax between 1000 and 51. 0: peptides inducing no immune response or a very low immune response with OD halfmax around 50 or lower.			

Claims

- Composition comprising at least one peptide or polypeptide comprising the amino acid sequence



wherein

X_1 is any amino acid residue,

X_2 is an amino acid residue selected from the group consisting of lysine (K), arginine (R), alanine (A) and histidine (H),

X_3 is an amino acid residue selected from the group consisting of asparagine (N), glutamine (Q), serine (S), glycine (G) and alanine (A), preferably asparagine (N), serine (S), glycine (G) and alanine (A),

X_4 is an amino acid residue selected from the group consisting of glutamic acid (E), aspartic acid (D) and alanine (A), X_5 is an amino acid residue selected from the group consisting of glutamic acid (E) and aspartic acid (D),

X_6 is an amino acid residue selected from the group consisting of alanine (A) and tyrosine (Y),

X_7 is any amino acid residue,

n and m , independently, are 0 or an integer of more than 0,

and wherein the amino acid sequence according to Formula I is not identical with, or does not comprise the 7-mer polypeptide fragment of alpha-synuclein having the amino acid sequence KNEEGAP,

said at least one peptide or polypeptide having a binding capacity to an antibody which is specific for an epitope of alpha-synuclein comprising the amino acid sequence KNEEGAP for use in preventing and/or treating synucleinopathies.

2. Composition for the use according to claim 1, wherein X_2 is an amino acid residue selected from the group consisting of lysine (K) and arginine (R) and/or X_6 is alanine (A).
3. Composition for the use according to claim 1 or 2 **characterised in that** the peptide or polypeptide comprises an amino acid sequence selected from the group consisting of $(X_1)_n$ KNDEGAP(X_7) $_m$, $(X_1)_n$ ANEEGAP(X_7) $_m$, $(X_1)_n$ KAEEGAP(X_7) $_m$, $(X_1)_n$ KNAEGAP(X_7) $_m$, $(X_1)_n$ RNEEGAP(X_7) $_m$, $(X_1)_n$ HNEEGAP(X_7) $_m$, $(X_1)_n$ KNEDGAP(X_7) $_m$, $(X_1)_n$ KQEEGAP(X_7) $_m$, $(X_1)_n$ KSEEGAP(X_7) $_m$, $(X_1)_n$ KNDDGAP(X_7) $_m$, $(X_1)_n$ RNDEGAP(X_7) $_m$, $(X_1)_n$ RNEDGAP(X_7) $_m$, $(X_1)_n$ RQEEGAP(X_7) $_m$, $(X_1)_n$ RSEEGAP(X_7) $_m$, $(X_1)_n$ ANDEGAP(X_7) $_m$, $(X_1)_n$ ANEDGAP(X_7) $_m$, $(X_1)_n$ HSEEGAP(X_7) $_m$, $(X_1)_n$ ASEEGAP(X_7) $_m$, $(X_1)_n$ HNEDGAP(X_7) $_m$, $(X_1)_n$ HNDEGAP(X_7) $_m$, $(X_1)_n$ RNAEGAP(X_7) $_m$, $(X_1)_n$ HNAEGAP(X_7) $_m$, $(X_1)_n$ KSAEGAP(X_7) $_m$, $(X_1)_n$ KSDEGAP(X_7) $_m$, $(X_1)_n$ KSEDGAP(X_7) $_m$, $(X_1)_n$ RQDEGAP(X_7) $_m$, $(X_1)_n$ RQEDGAP(X_7) $_m$, $(X_1)_n$ HSAEGAP(X_7) $_m$, $(X_1)_n$ RSAEGAP(X_7) $_m$, $(X_1)_n$ RSDEGAP(X_7) $_m$, $(X_1)_n$ RSEDGAP(X_7) $_m$, $(X_1)_n$ HSDEGAP(X_7) $_m$, $(X_1)_n$ HSEDGAP(X_7) $_m$, $(X_1)_n$ RQDDGAP(X_7) $_m$, preferably $(X_1)_n$ KNDEGAP(X_2) $_m$, $(X_1)_n$ RNEEGAP(X_2) $_m$, $(X_1)_n$ RNDEGAP(X_2) $_m$, $(X_1)_n$ KNAEGAP(X_2) $_m$, $(X_1)_n$ KSDEGAP(X_2) $_m$, $(X_1)_n$ RNAEGAP(X_2) $_m$ or $(X_1)_n$ RSEEGAP(X_2) $_m$.
4. Composition for the use according to any one of claims 1 to 3, **characterised in that** n and/or m are 1 and X_1 and/or X_7 are cysteine (C).
5. Composition for the use according to any one of claims 1 to 4, **characterised in that** the peptide or polypeptide comprises 7 to 30, preferably 7 to 20, more preferably 7 to 16, most preferably 8, amino acid residues.
6. Composition for the use according to any one of claims 1 to 5, **characterised in that** the synucleinopathy is selected from the group consisting of Lewy Body Disorders (LBDs), preferably Parkinson's Disease (PD), Parkinson's Disease with Dementia (PDD) and Dementia with Lewy Bodies (DLB), as well as Multiple System Atrophy (MSA) or Neurodegeneration with Brain Iron Accumulation type I (NBIA Type I).
7. Composition for the use according to any one of claims 1 to 6, **characterised in that** the at least one peptide or polypeptide is coupled to a pharmaceutically acceptable carrier, preferably KLH (Keyhole Limpet Hemocyanin).
8. Composition for the use according to any one of claims 1 to 7, **characterised in that** the at least one peptide or polypeptide is formulated for intravenous, subcutaneous, intradermal or intramuscular administration.
9. Composition for the use according to any one of claims 1 to 8, **characterised in that** the at least one peptide or polypeptide is formulated with an adjuvant, preferably aluminium hydroxide.
10. Composition for the use according to any one of claims 1 to 9, **characterised in that** the at least one peptide or polypeptide is contained in an amount of from 0.1 ng to 10 mg, preferably 10 ng to 1 mg, in particular 100 ng to 100 μ g.
11. Peptide having an amino acid sequence selected from the group consisting of $(X_1)_n$ KNDEGAP(X_7) $_m$, $(X_1)_n$ ANEEGAP(X_7) $_m$, $(X_1)_n$ KAEEGAP(X_7) $_m$, $(X_1)_n$ KNAEGAP(X_7) $_m$, $(X_1)_n$ RNEEGAP(X_7) $_m$, $(X_1)_n$ HNEEGAP(X_7) $_m$,

(X₁)_nKNEDGAP(X₇)_m, (X₁)_nKQEEGAP(X₇)_m, (X₁)_nKSEEGAP(X₇)_m, (X₁)_nKNDDGAP(X₇)_m, (X₁)_nRNDEGAP(X₇)_m, (X₁)_nRNEDGAP(X₇)_m, (X₁)_nRQEEGAP(X₇)_m, (X₁)_nRSEEGAP(X₇)_m, (X₁)_nANDEGAP(X₇)_m, (X₁)_nANEDGAP(X₇)_m, (X₁)_nHSEEGAP(X₇)_m, (X₁)_nASEEGAP(X₇)_m, (X₁)_nHNEDGAP(X₇)_m, (X₁)_nHNDEGAP(X₇)_m, (X₁)_nRNAEGAP(X₇)_m, (X₁)_nHNAEGAP(X₇)_m, (X₁)_nKSAEGAP(X₇)_m, (X₁)_nKSDEGAP(X₇)_m, (X₁)_nKSEDEGAP(X₇)_m, (X₁)_nRQDEGAP(X₇)_m, (X₁)_nRQEDGAP(X₇)_m, (X₁)_nHSAEGAP(X₇)_m, (X₁)_nRSAEGAP(X₇)_m, (X₁)_nRSDEGAP(X₇)_m, (X₁)_nRSEDEGAP(X₇)_m, (X₁)_nHSDEGAP(X₇)_m, (X₁)_nHSEDEGAP(X₇)_m and (X₁)_nRQDDGAP(X₇)_m, wherein X₁ and X₇ is cysteine and n and m, independently, are 0 or 1.

12. Peptide according to claim 11, **characterised in that** the peptide is coupled to a pharmaceutically acceptable carrier, preferably KLH (Keyhole Limpet Hemocyanin).

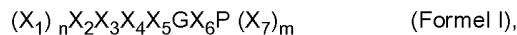
13. Peptide according to claim 11 or 12 for use in preventing and/or treating synucleinopathies.

14. Pharmaceutical formulation comprising at least one peptide according to claim 11 or 12.

15. Pharmaceutical formulation according to claim 14 for use as a vaccine.

Patentansprüche

1. Zusammensetzung, umfassend mindestens ein Peptid oder Polypeptid, das die Aminosäuresequenz



umfasst, wobei

X₁ für einen beliebigen Aminosäurerest steht,

X₂ für einen Aminosäurerest steht, der ausgewählt ist aus der Gruppe bestehend aus Lysin (K), Arginin (R), Alanin (A) und Histidin (H),

X₃ für einen Aminosäurerest steht, der ausgewählt ist aus der Gruppe bestehend aus Asparagin (N), Glutamin (Q), Serin (S), Glycin (G) und Alanin (A), bevorzugt für Asparagin (N), Serin (S), Glycin (G) und Alanin (A),

X₄ für einen Aminosäurerest steht, der ausgewählt ist aus der Gruppe bestehend aus Glutaminsäure (E), Asparaginsäure (D) und Alanin (A),

X₅ für einen Aminosäurerest steht, der ausgewählt ist aus der Gruppe bestehend aus Glutaminsäure (E) und Asparaginsäure (D),

X₆ für einen Aminosäurerest steht, der ausgewählt ist aus der Gruppe bestehend aus Alanin (A) und Tyrosin (Y),

X₇ für einen beliebigen Aminosäurerest steht,

n und m jeweils unabhängig voneinander für 0 oder eine ganze Zahl größer 0 stehen,

und wobei die Aminosäuresequenz gemäß Formel I nicht mit dem 7-meren Polypeptidfragment von Alpha-Synuklein mit der Aminosäuresequenz KNEEGAP identisch ist bzw. dieses nicht umfasst,

wobei das mindestens eine Peptid oder Polypeptid eine Bindungskapazität gegenüber einem Antikörper aufweist, der für ein Epitop von Alpha-Synuklein, das die Aminosäuresequenz KNEEGAP umfasst, spezifisch ist, zur Verwendung bei der Prävention und/oder Behandlung von Synukleinopathien.

2. Zusammensetzung zur Verwendung nach Anspruch 1, wobei X₂ für einen Aminosäurerest steht, der ausgewählt ist aus der Gruppe bestehend aus Lysin (K) und Arginin (R) und/oder X₆ für Alanin (A) steht.

3. Zusammensetzung zur Verwendung nach Anspruch 1 oder 2, **dadurch gekennzeichnet, dass** das Peptid oder Polypeptid eine Aminosäuresequenz umfasst, die ausgewählt ist aus der Gruppe bestehend aus (X₁)_nKNDEGAP(X₇)_m, (X₁)_nANEEGAP(X₇)_m, (X₁)_nKAEEGAP(X₇)_m, (X₁)_nKNAEGAP(X₇)_m, (X₁)_nRNEEGAP(X₇)_m, (X₁)_nHNEEGAP(X₇)_m, (X₁)_nKNEDGAP(X₇)_m, (X₁)_nKQEEGAP(X₇)_m, (X₁)_nKSEEGAP(X₇)_m, (X₁)_nKNDDGAP(X₇)_m, (X₁)_nRNDEGAP(X₇)_m, (X₁)_nRNEDGAP(X₇)_m, (X₁)_nRQEEGAP(X₇)_m, (X₁)_nRSEEGAP(X₇)_m, (X₁)_nANDEGAP(X₇)_m, (X₁)_nANEDGAP(X₇)_m, (X₁)_nHSEEGAP(X₇)_m, (X₁)_nASEEGAP(X₇)_m, (X₁)_nHNEDGAP(X₇)_m, (X₁)_nHNDEGAP(X₇)_m, (X₁)_nRNAEGAP(X₇)_m, (X₁)_nHNAEGAP(X₇)_m, (X₁)_nKSAEGAP(X₇)_m, (X₁)_nKSDEGAP(X₇)_m, (X₁)_nKSEDEGAP(X₇)_m, (X₁)_nRQDEGAP(X₇)_m, (X₁)_nRQEDGAP(X₇)_m, (X₁)_nHSAEGAP(X₇)_m, (X₁)_nRSAEGAP(X₇)_m, (X₁)_nRSDEGAP(X₇)_m, (X₁)_nRSEDEGAP(X₇)_m, (X₁)_nHSDEGAP(X₇)_m, (X₁)_nHSEDEGAP(X₇)_m, (X₁)_nRQDDGAP(X₇)_m, bevorzugt (X₁)_nKNDEGAP(X₂)_m, (X₁)_nRNEEGAP(X₂)_m, (X₁)_nRNDEGAP(X₂)_m, (X₁)_nKNAEGAP(X₂)_m, (X₁)_nKSDEGAP(X₂)_m, (X₁)_nRNA-

EGAP(X₂)_m oder (X₁)_nRSEEGAP(X₂)_m.

4. Zusammensetzung zur Verwendung nach einem der Ansprüche 1 bis 3, **dadurch gekennzeichnet, dass** jeweils n und/oder m für 1 und X₁ und/oder X₇ für Cystein (C) stehen.

5. Zusammensetzung zur Verwendung nach einem der Ansprüche 1 bis 4, **dadurch gekennzeichnet, dass** das Peptid oder Polypeptid 7 bis 30, bevorzugt 7 bis 20, mehr bevorzugt 7 bis 16 und am meisten bevorzugt 8 Aminosäurereste umfasst.

6. Zusammensetzung zur Verwendung nach einem der Ansprüche 1 bis 5, **dadurch gekennzeichnet, dass** die Synukleinopathie ausgewählt ist aus der Gruppe bestehend aus Lewy-Body-Erkrankungen (LBDs), bevorzugt Parkinson-Krankheit (PK), Parkinson-Krankheit mit Demenz (PKD) und Demenz mit Lewy-Bodies (DLB), sowie Multisystematrophie (MSA) oder Neurodegeneration mit Eisenablagerung im Gehirn des Typ I (NBIA-Typ I).

7. Verwendung nach einem der Ansprüche 1 bis 6, **dadurch gekennzeichnet, dass** das mindestens eine Peptid oder Polypeptid an einen pharmazeutisch akzeptablen Träger, vorzugsweise KLH (Schlüssellockschnecken-Hämocyanin) gebunden ist.

8. Zusammensetzung zur Verwendung nach einem der Ansprüche 1 bis 7, **dadurch gekennzeichnet, dass** das mindestens eine Peptid oder Polypeptid zur intravenösen, subkutanen, intradermalen oder intramuskulären Verabreichung formuliert ist.

9. Zusammensetzung zur Verwendung nach einem der Ansprüche 1 bis 8, **dadurch gekennzeichnet, dass** das mindestens eine Peptid oder Polypeptid mit einem Adjuvans, vorzugsweise Aluminiumhydroxid, formuliert ist.

10. Zusammensetzung zur Verwendung nach einem der Ansprüche 1 bis 9, **dadurch gekennzeichnet, dass** das mindestens eine Peptid oder Polypeptid in einer Menge von 0,1 ng bis 10 mg, bevorzugt von 10 ng bis 1 mg, insbesondere von 100 ng bis 100 µg, enthalten ist.

11. Peptid mit einer Aminosäuresequenz, die ausgewählt ist aus der Gruppe bestehend aus (X₁)_nKNDEGAP(X₇)_m, (X₁)_nANEEGAP(X₇)_m, (X₁)_nKAEEGAP(X₇)_m, (X₁)_nKNAEGAP(X₇)_m, (X₁)_nRNEEGAP(X₇)_m, (X₁)_nHNEEGAP(X₇)_m, (X₁)_nKNEDGAP(X₇)_m, (X₁)_nKQEEGAP(X₇)_m, (X₁)_nKSEEGAP(X₇)_m, (X₁)_nKNDDGAP(X₇)_m, (X₁)_nRNDEGAP(X₇)_m, (X₁)_nRNEDGAP(X₇)_m, (X₁)_nRQEEGAP(X₇)_m, (X₁)_nRSEEGAP(X₇)_m, (X₁)_nANDEGAP(X₇)_m, (X₁)_nANEDGAP(X₇)_m, (X₁)_nHSEEGAP(X₇)_m, (X₁)_nASEEGAP(X₇)_m, (X₁)_nHNEDGAP(X₇)_m, (X₁)_nHNDEGAP(X₇)_m, (X₁)_nRNAEGAP(X₇)_m, (X₁)_nHNAEGAP(X₇)_m, (X₁)_nKSAEGAP(X₇)_m, (X₁)_nKSDEGAP(X₇)_m, (X₁)_nKSEDEGAP(X₇)_m, (X₁)_nRQDEGAP(X₇)_m, (X₁)_nRQEDGAP(X₇)_m, (X₁)_nHSAEGAP(X₇)_m, (X₁)_nRSAEGAP(X₇)_m, (X₁)_nRSDEGAP(X₇)_m, (X₁)_nRSDEGAP(X₇)_m, (X₁)_nHSDEGAP(X₇)_m, (X₁)_nHSEDEGAP(X₇)_m und (X₁)_nRQDDGAP(X₇)_m, wobei jeweils X₁ und X₇ für Cystein und n und m unabhängig voneinander für 0 oder 1 stehen.

12. Peptid nach Anspruch 11, **dadurch gekennzeichnet, dass** das Peptid an einen pharmazeutisch akzeptablen Träger, bevorzugt KLH (Schlüssellockschnecken-Hämocyanin), gekoppelt ist.

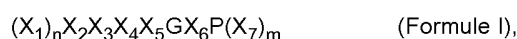
13. Peptid nach Anspruch 11 oder 12 zur Verwendung bei der Prävention und/oder Behandlung von Synukleinopathien.

14. Pharmazeutische Formulierung, umfassend mindestens ein Peptid nach Anspruch 11 oder 12.

15. Pharmazeutische Formulierung nach Anspruch 14 zur Verwendung als Impfstoff.

Revendications

1. Composition comprenant au moins un peptide ou polypeptide comprenant la séquence d'acides aminés



dans laquelle

X₁ est un quelconque résidu d'acide aminé,

X_2 est un résidu d'acide aminé sélectionné dans le groupe consistant en la lysine (K), l'arginine (R), l'alanine (A) et l'histidine (H),

X_3 est un résidu d'acide aminé sélectionné dans le groupe consistant en l'asparagine (N), la glutamine (Q), la sérine (S), la glycine (G) et l'alanine (A), de préférence l'asparagine (N), la sérine (S), la glycine (G) et l'alanine (A),

X_4 est un résidu d'acide aminé sélectionné dans le groupe consistant en l'acide glutamique (E), l'acide aspartique (D) et l'alanine (A),

X_5 est un résidu d'acide aminé sélectionné dans le groupe consistant en l'acide glutamique (E) et l'acide aspartique (D),

X_6 est un résidu d'acide aminé sélectionné dans le groupe consistant en l'alanine (A) et la tyrosine (Y),

X_7 est un quelconque résidu d'acide aminé,

n et m, indépendamment, sont 0 ou un nombre entier supérieur à 0,

et où la séquence d'acides aminés selon la Formule I n'est pas identique à, ou ne comprend pas le fragment polypeptidique 7-mère de l'alpha-synucléine ayant la séquence d'acides aminés KNEEGAP,

ledit au moins un peptide ou polypeptide possédant une capacité de liaison à un anticorps qui est spécifique pour un épitope de l'alpha-synucléine comprenant la séquence d'acides aminés KNEEGAP destinée à être utilisée dans la prévention et/ou le traitement de synucléinopathies.

2. Composition destinée à être utilisée selon la revendication 1, dans laquelle X_2 est un résidu d'acide aminé sélectionné dans le groupe consistant en la lysine (K) et l'arginine (R) et/ou X_6 est l'alanine (A).

3. Composition destinée à être utilisée selon la revendication 1 ou 2, **caractérisée en ce que** le peptide ou polypeptide comprend une séquence d'acides aminés sélectionnée dans le groupe consistant en $(X_1)_n$ KNDEGAP(X_7)_m, $(X_1)_n$ ANEEGAP(X_7)_m, $(X_1)_n$ KAEEGAP(X_7)_m, $(X_1)_n$ KNAEGAP(X_7)_m, $(X_1)_n$ RNEEGAP(X_7)_m, $(X_1)_n$ HNEEGAP(X_7)_m, $(X_1)_n$ KNEDGAP(X_7)_m, $(X_1)_n$ KQEEGAP(X_7)_m, $(X_1)_n$ KSEEGAP(X_7)_m, $(X_1)_n$ KNDDGAP(X_7)_m, $(X_1)_n$ RNDEGAP(X_7)_m, $(X_1)_n$ RNEDGAP(X_7)_m, $(X_1)_n$ RQEEGAP(X_7)_m, $(X_1)_n$ RSEEGAP(X_7)_m, $(X_1)_n$ ANDEGAP(X_7)_m, $(X_1)_n$ ANEDGAP(X_7)_m, $(X_1)_n$ HSEEGAP(X_7)_m, $(X_1)_n$ ASEEGAP(X_7)_m, $(X_1)_n$ HNEDGAP(X_7)_m, $(X_1)_n$ HNDEGAP(X_7)_m, $(X_1)_n$ RNAEGAP(X_7)_m, $(X_1)_n$ HNAEGAP(X_7)_m, $(X_1)_n$ KSAEGAP(X_7)_m, $(X_1)_n$ KSDEGAP(X_7)_m, $(X_1)_n$ KSEEGAP(X_7)_m, $(X_1)_n$ RQDEGAP(X_7)_m, $(X_1)_n$ RQEDGAP(X_7)_m, $(X_1)_n$ HSAEGAP(X_7)_m, $(X_1)_n$ RSAEGAP(X_7)_m, $(X_1)_n$ RSDEGAP(X_7)_m, $(X_1)_n$ RSEDGAP(X_7)_m, $(X_1)_n$ HSDEGAP(X_7)_m, $(X_1)_n$ HSEDGAP(X_7)_m, $(X_1)_n$ RQDDGAP(X_7)_m, de préférence $(X_1)_n$ KNDEGAP(X_2)_m, $(X_1)_n$ RNEEGAP(X_2)_m, $(X_1)_n$ RNDEGAP(X_2)_m, $(X_1)_n$ KNAEGAP(X_2)_m, $(X_1)_n$ KSDEGAP(X_2)_m, $(X_1)_n$ RNAEGAP(X_2)_m ou $(X_1)_n$ RSEEGAP(X_2)_m.

4. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 3, **caractérisée en ce que** n et/ou m sont 1 et X_1 et/ou X_7 sont la cystéine (C).

5. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 4, **caractérisée en ce que** le peptide ou polypeptide comprend 7 à 30, de préférence 7 à 20, de manière davantage préférée 7 à 16, de la manière la plus préférée entre toutes 8, résidus d'acides aminés.

6. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 5, **caractérisée en ce que** la synucléinopathie est sélectionnée dans le groupe consistant en des maladies à corps de Lewy (MCL), de préférence la maladie de Parkinson (MP), la *démence* de la maladie de Parkinson (DMP) et la démence à corps de Lewy (DCL), ainsi que l'atrophie multisystématisée (AMS) ou la neurodégénérescence avec accumulation de fer dans le cerveau de type I (NBIA de type I).

7. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 6, **caractérisée en ce que** le au moins un peptide ou polypeptide est couplé à un véhicule pharmaceutiquement acceptable, de préférence la KLH (hémocyanine de patelle).

8. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 7, **caractérisée en ce que** le au moins un peptide ou polypeptide est formulé pour une administration intraveineuse, sous-cutanée, intradermique ou intramusculaire.

9. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 8, **caractérisée en ce que** le au moins un peptide ou polypeptide est formulé avec un adjuvant, de préférence l'hydroxyde d'aluminium.

10. Composition destinée à être utilisée selon l'une quelconque des revendications 1 à 9, **caractérisée en ce que** le

au moins un peptide ou polypeptide est contenu en une quantité comprise entre 0,1 ng et 10 mg, de préférence 10 ng et 1 mg, en particulier 100 ng et 100 µg.

11. Peptide ayant une séquence d'acides aminés sélectionnée dans le groupe consistant en $(X_1)_n\text{KNDEGAP}(X_7)_m$, $(X_1)_n\text{ANEEGAP}(X_7)_m$, $(X_1)_n\text{KAEEGAP}(X_7)_m$, $(X_1)_n\text{KNAEGAP}(X_7)_m$, $(X_1)_n\text{RNEEGAP}(X_7)_m$, $(X_1)_n\text{HNEEGAP}(X_7)_m$, $(X_1)_n\text{KNEDGAP}(X_7)_m$, $(X_1)_n\text{KQEEGAP}(X_7)_m$, $(X_1)_n\text{KSEEGAP}(X_7)_m$, $(X_1)_n\text{KNDDGAP}(X_7)_m$, $(X_1)_n\text{RNDEGAP}(X_7)_m$, $(X_1)_n\text{RNEDGAP}(X_7)_m$, $(X_1)_n\text{RQEEGAP}(X_7)_m$, $(X_1)_n\text{RSEEGAP}(X_7)_m$, $(X_1)_n\text{ANDEGAP}(X_7)_m$, $(X_1)_n\text{ANEDGAP}(X_7)_m$, $(X_1)_n\text{HSEEGAP}(X_7)_m$, $(X_1)_n\text{ASEEGAP}(X_7)_m$, $(X_1)_n\text{HNEDGAP}(X_7)_m$, $(X_1)_n\text{HNDEGAP}(X_7)_m$, $(X_1)_n\text{RNAEGAP}(X_7)_m$, $(X_1)_n\text{HNAEGAP}(X_7)_m$, $(X_1)_n\text{KSAEGAP}(X_7)_m$, $(X_1)_n\text{KSDEGAP}(X_7)_m$, $(X_1)_n\text{KSEDGAP}(X_7)_m$, $(X_1)_n\text{RQDEGAP}(X_7)_m$, $(X_1)_n\text{RQEDGAP}(X_7)_m$, $(X_1)_n\text{HSAEGAP}(X_7)_m$, $(X_1)_n\text{RSAEGAP}(X_7)_m$, $(X_1)_n\text{RSDEGAP}(X_7)_m$, $(X_1)_n\text{RSEDGAP}(X_7)_m$, $(X_1)_n\text{HSDEGAP}(X_7)_m$, $(X_1)_n\text{HSEDGAP}(X_7)_m$, $(X_1)_n\text{RQDDGAP}(X_7)_m$, dans laquelle X_1 et X_7 sont la cystéine et n et m, indépendamment, sont 0 ou 1.

12. Peptide selon la revendication 11, **caractérisé en ce que** le peptide est couplé à un véhicule pharmaceutiquement acceptable, de préférence la KLH (hémocyanine de patelle).

13. Peptide selon la revendication 11 ou 12 destiné à être utilisé dans la prévention et/ou le traitement de synucléinopathies.

14. Formulation pharmaceutique comprenant au moins un peptide selon la revendication 11 ou 12.

15. Formulation pharmaceutique selon la revendication 14 destinée à être utilisée en tant que vaccin.

Alpha-synuclein protein sequence:

10	20	30	40	50	60
MDVFMKGLSK	AKEGVVAAAE	GTKQGVAAEA	GKTKEGVLYV	GSKTKEGVVH	GVATVAEKT
70	80	90	100	110	120
EQVTNVGGAV	VTGVTAVAQK	TVEGAGSIAA	ATGEVKKDQL	<u>GKNEEGAPQE</u>	GILEDMPVDP
130	140				
DNEAYEMPSE	EGYQDYEP				

Fig. 1

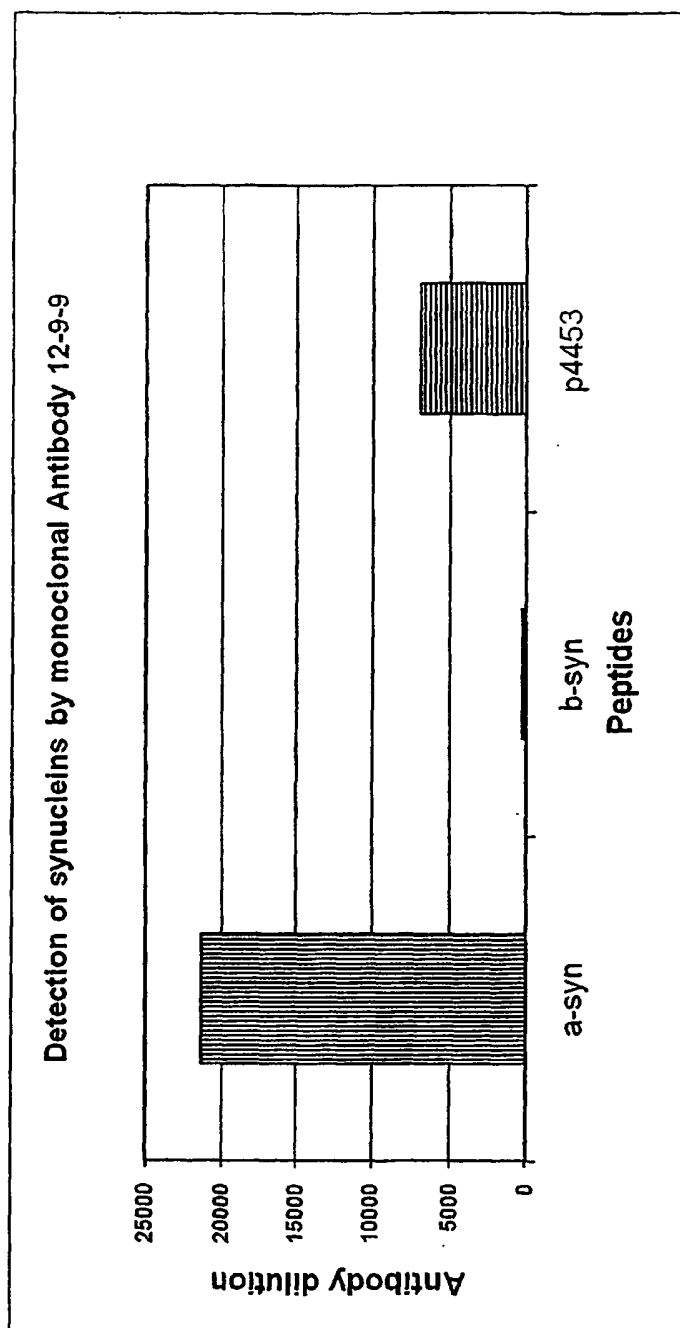


Fig. 2

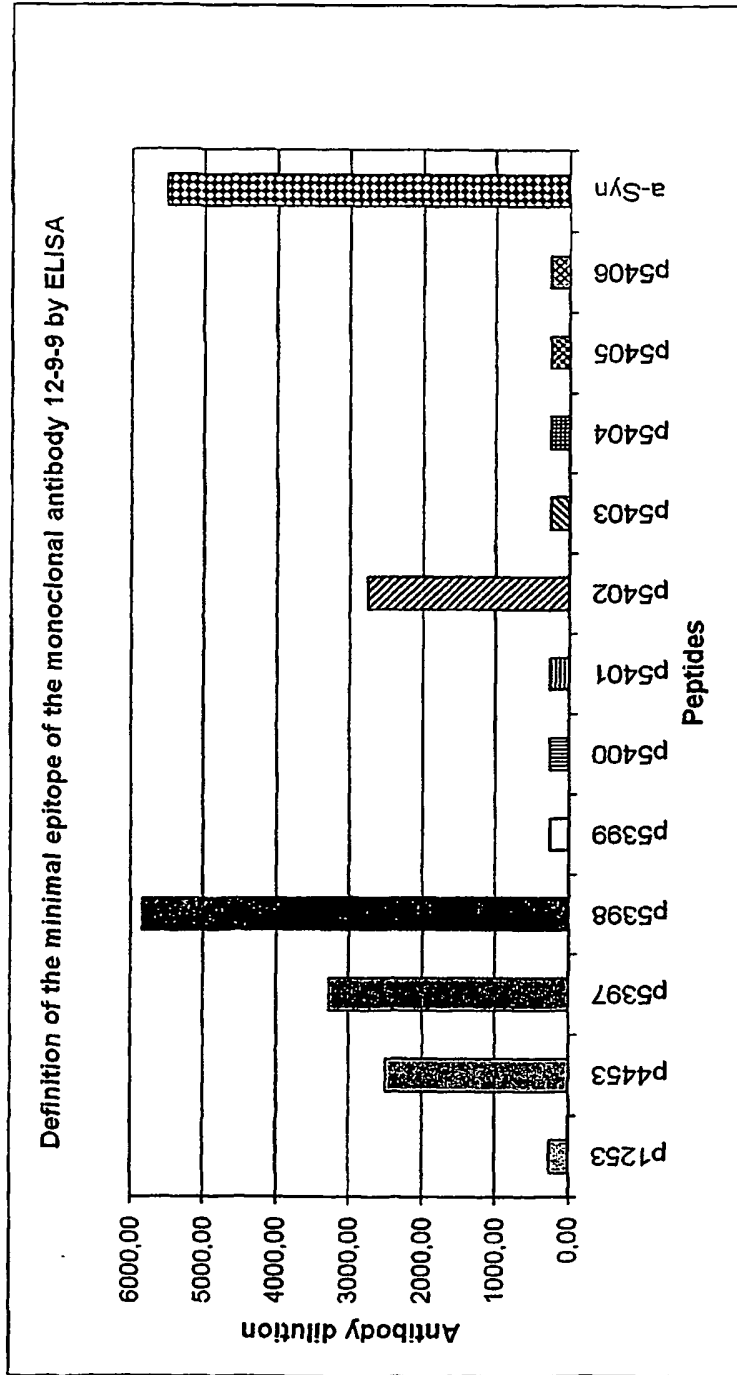


Fig. 3

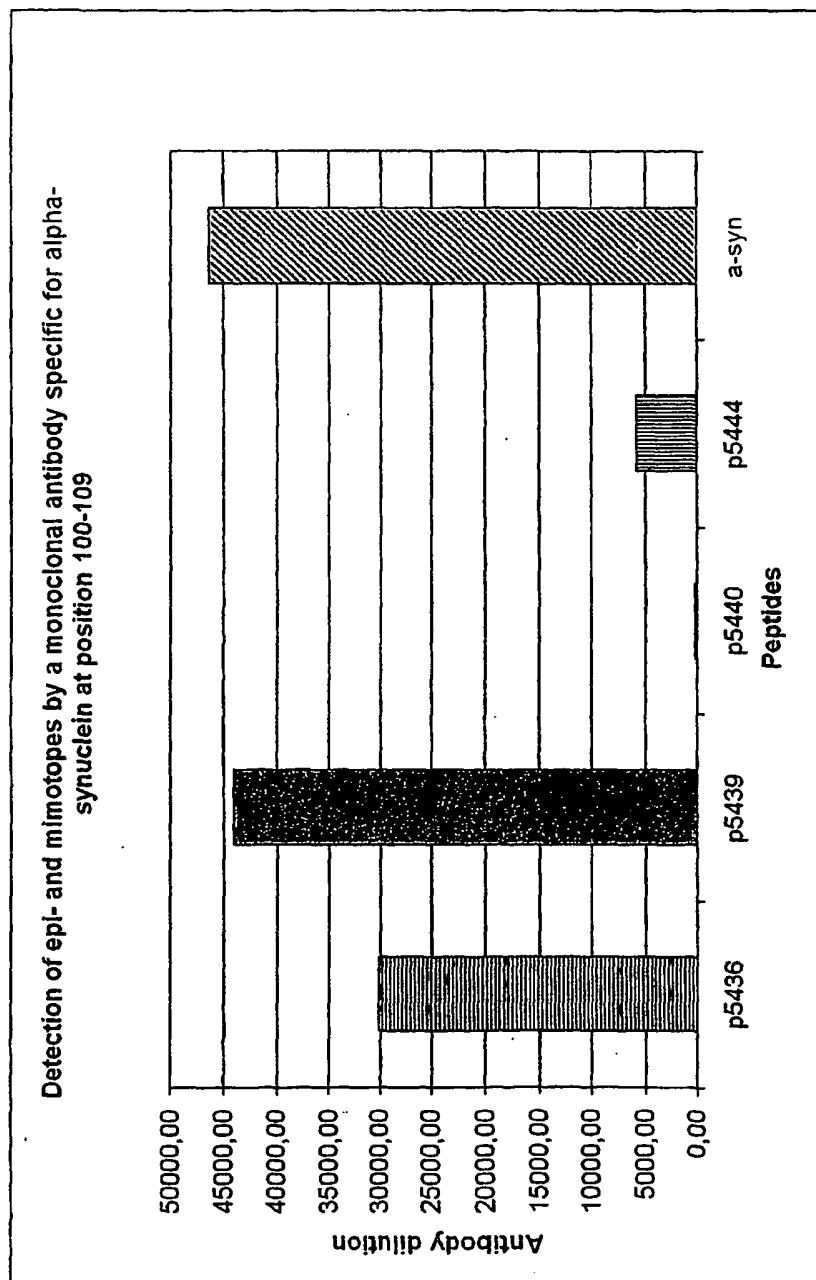


Fig. 4

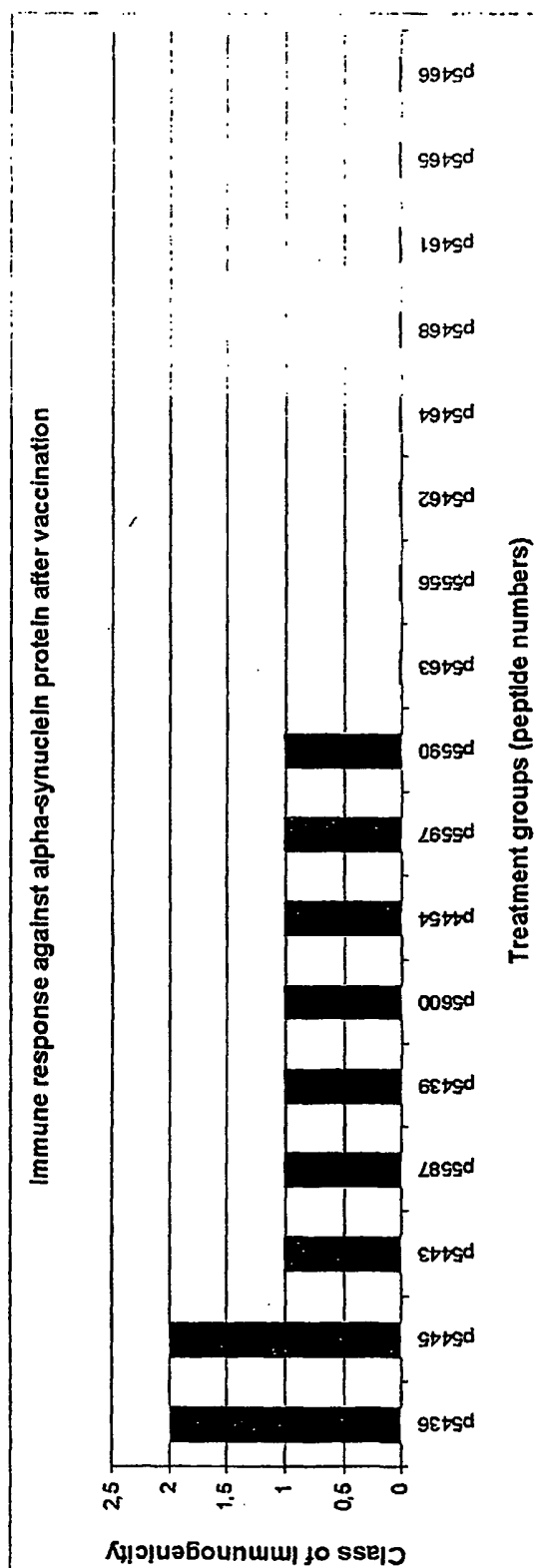


Fig. 5

REFERENCES CITED IN THE DESCRIPTION

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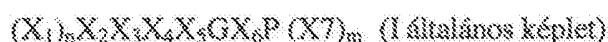
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Alfa-synuclein epitopok mimotópjainak alkalmazása Lewy-test betegségek kezelésében

SZABADALMI IGÉNYPONTOK

1. Készítmény, amely legalább egy peptidet, vagy polipeptidet tartalmaz, amely a következő aminosav szekvenciát tartalmazza:



X_1 jelentése bármilyen aminosav csoport,

X_2 jelentése aminosav csoport, amelyet a következő csoportból választhatunk ki: lizin (K), arginin (R), és hisztidin (H),

X_3 jelentése aminosav csoport, amelyet a következő csoportból választhatunk ki: aszparagin (N), glutamin (Q), szerin (S), glicin (G) és alanin (A), előnyösen aszparagin (N), szerin (S), glicin (G) és alanin (A),

X_4 jelentése aminosav csoport, amelyet a következő csoportból választhatunk ki: glutaminsav (E), aszparaginsav (D) és alanin (A),

X_5 jelentése aminosav csoport, amelyet a következő csoportból választhatunk ki: glutaminsav (E) és aszparaginsav (D),

X_6 jelentése aminosav csoport, amelyet a következő csoportból választhatunk ki: alanin (A) és tirozin (Y),

X_7 jelentése bármilyen aminosav csoport,

n és m értéke egymástól függetlenül 0, vagy 0-nál nagyobb egész szám,

és ahol a (I) általános képlet szerinti aminosav szekvencia nem azonos az alfa-synuclein héttagú, a KNEEGAP aminosav szekvenciával rendelkező polipeptid-fragmensével, vagy nem tartalmazza azt, a szóban forgó legalább egy peptid vagy polipeptid kötési kapacitással rendelkezik egy ellenanyaghoz, amely specifikus az alfa-synucleinnek a KNEEGAP aminosav szekvenciát tartalmazó egyik epitopjára

synucleinopátiák megelőzésében és/vagy kezelésében történő alkalmazásra.

2. Az 1. igénypont szerinti készítmény alkalmazása, ahol X_2 jelentése aminosav csoport, amelyet a következő csoportból választhatunk ki: lizin (K), és arginin (R), és/vagy X_6 jelentése alanin (A).

3. Az 1. vagy 2. igénypont szerinti készítmény alkalmazása, ahol a peptid vagy polipeptid tartalmaz egy, az alábbi csoportból választható aminosav szekvenciát: $(X_1)_n \text{KNDEGAP}(X_7)_m$, $(X_1)_n \text{ANEEGAP}(X_7)_m$, $(X_1)_n \text{KAEAGAP}(X_7)_m$, $(X_1)_n \text{KNAEGAP}(X_7)_m$, $(X_1)_n \text{RNEEGAP}(X_7)_m$,

$(X_1)_n\text{HNEEGAP}(X_7)_m$, $(X_1)_n\text{KNEDGAP}(X_7)_m$, $(X_1)_n\text{KQEEGAP}(X_7)_m$, $(X_1)_n\text{KSEEGAP}(X_7)_m$,
 $(X_1)_n\text{KNDDGAP}(X_7)_m$, $(X_1)_n\text{RNDEGAP}(X_7)_m$, $(X_1)_n\text{RNEDGAP}(X_7)_m$, $(X_1)_n\text{RQEEGAP}(X_7)_m$,
 $(X_1)_n\text{RSEEGAP}(X_7)_m$, $(X_1)_n\text{ANDEGAP}(X_7)_m$, $(X_1)_n\text{ANEDGAP}(X_7)_m$, $(X_1)_n\text{HSEEGAP}(X_7)_m$,
 $(X_1)_n\text{ASEEGAP}(X_7)_m$, $(X_1)_n\text{HNEDGAP}(X_7)_m$, $(X_1)_n\text{HNDEGAP}(X_7)_m$, $(X_1)_n\text{RNAEGAP}(X_7)_m$,
 $(X_1)_n\text{HNAEGAP}(X_7)_m$, $(X_1)_n\text{KSAEGAP}(X_7)_m$, $(X_1)_n\text{KSDEGAP}(X_7)_m$, $(X_1)_n\text{KSEDGAP}(X_7)_m$,
 $(X_1)_n\text{RQDEGAP}(X_7)_m$, $(X_1)_n\text{RQEDGAP}(X_7)_m$, $(X_1)_n\text{HSAEGAP}(X_7)_m$, $(X_1)_n\text{RSAEGAP}(X_7)_m$,
 $(X_1)_n\text{RSDEGAP}(X_7)_m$, $(X_1)_n\text{RSEDGAP}(X_7)_m$, $(X_1)_n\text{HSDEGAP}(X_7)_m$, $(X_1)_n\text{HSEDGAP}(X_7)_m$,
 $(X_1)_n\text{RQDDGAP}(X_7)_m$, előnyösen $(X_1)_n\text{KNDEGAP}(X_2)_m$, $(X_1)_n\text{RNEEGAP}(X_2)_m$,
 $(X_1)_n\text{RNDEGAP}(X_2)_m$, $(X_1)_n\text{KNAEGAP}(X_2)_m$, $(X_1)_n\text{KSDEGAP}(X_2)_m$, $(X_1)_n\text{RNAEGAP}(X_2)_m$ vagy
 $(X_1)_n\text{RSEEGAP}(X_2)_m$.

4. Az 1-3. igénypontok bármelyike szerinti készítmény alkalmazása, ahol n és/vagy m értéke 1, és X_1 és/vagy X_7 jelentése cisztein (C)

5. Az 1-4. igénypontok bármelyike szerinti készítmény alkalmazása, ahol a peptid vagy polipeptid 7-30, előnyösen 7-20, előnyösebben 7-16, legelőnyösebben 8 aminosav csoportot tartalmaz.

6. Az 1-5. igénypontok bármelyike szerinti készítmény alkalmazása, ahol a synucleinopátiát a következő csoportból választhatjuk ki: Lewy test rendellenességek (LBD-k), előnyösen Parkinson kór (PD), Parkinson kór demenciával (PDD) és Demencia Lewy testekkel (DLB), valamint Multiszisztémás atrófia (MSA) vagy Neurodegeneráció I-es típusú agyi vas akkumulációval (NBIA I-es típus).

7. Az 1-6. igénypontok bármelyike szerinti készítmény alkalmazása, ahol legalább egy peptid vagy polipeptid egy gyógyászatilag elfogadható hordozóhoz, előnyösen KLH-hoz (fűrócsiga hemocianin) kapcsolódik.

8. Az 1-7. igénypontok bármelyike szerinti készítmény alkalmazása, ahol legalább egy peptid vagy polipeptid intravénás, szubkután, intradermális vagy intramuszkuláris beadáshoz van kisserelve.

9. Az 1-8. igénypontok bármelyike szerinti készítmény alkalmazása, ahol legalább egy peptid vagy polipeptid egy adjuvánssal, előnyösen alumínium-hidroxiddal van kisserelve.

10. Az 1-8. igénypontok bármelyike szerinti készítmény alkalmazása, ahol legalább egy peptid vagy polipeptid 0,1 ng – 10 mg, előnyösen 10 ng – 1 mg, pontosabban 100ng – 100 µg mennyiségben van jelen.

11. Peptid, amelynek az aminosav szekvenciáját a következő csoportból választhatjuk ki:
 $(X_1)_n\text{ANEEGAP}(X_7)_m$, $(X_1)_n\text{KAEEGAP}(X_7)_m$, $(X_1)_n\text{KNAEGAP}(X_7)_m$, $(X_1)_n\text{RNEEGAP}(X_7)_m$,
 $(X_1)_n\text{HNEEGAP}(X_7)_m$, $(X_1)_n\text{KNEDGAP}(X_7)_m$, $(X_1)_n\text{KQEEGAP}(X_7)_m$, $(X_1)_n\text{KSEEGAP}(X_7)_m$,

$(X_1)_n\text{KNDDGAP}(X_7)_m$, $(X_1)_n\text{RNDEGAP}(X_7)_m$, $(X_1)_n\text{RNEDGAP}(X_7)_m$, $(X_1)_n\text{RQEEGAP}(X_7)_m$,
 $(X_1)_n\text{RSEEGAP}(X_7)_m$, $(X_1)_n\text{ANDEGAP}(X_7)_m$, $(X_1)_n\text{ANEDGAP}(X_7)_m$, $(X_1)_n\text{HSEEGAP}(X_7)_m$,
 $(X_1)_n\text{ASEEGAP}(X_7)_m$, $(X_1)_n\text{HNEDGAP}(X_7)_m$, $(X_1)_n\text{HNDEGAP}(X_7)_m$, $(X_1)_n\text{RNAEGAP}(X_7)_m$,
 $(X_1)_n\text{HNAEGAP}(X_7)_m$, $(X_1)_n\text{KSAEGAP}(X_7)_m$, $(X_1)_n\text{KSDEGAP}(X_7)_m$, $(X_1)_n\text{KSEDGAP}(X_7)_m$,
 $(X_1)_n\text{RQDEGAP}(X_7)_m$, $(X_1)_n\text{RQEDGAP}(X_7)_m$, $(X_1)_n\text{HSAEGAP}(X_7)_m$, $(X_1)_n\text{RSAEGAP}(X_7)_m$,
 $(X_1)_n\text{RSDEGAP}(X_7)_m$, $(X_1)_n\text{RSEDGAP}(X_7)_m$, $(X_1)_n\text{HSDEGAP}(X_7)_m$, $(X_1)_n\text{HSEDGAP}(X_7)_m$,
 $(X_1)_n\text{RQDDGAP}(X_7)_m$, előnyösen $(X_1)_n\text{KNDEGAP}(X_2)_m$, $(X_1)_n\text{RNEEGAP}(X_2)_m$,
 $(X_1)_n\text{RNDEGAP}(X_2)_m$, $(X_1)_n\text{KNAEGAP}(X_2)_m$, $(X_1)_n\text{KSDEGAP}(X_2)_m$, $(X_1)_n\text{RNAEGAP}(X_2)_m$ vagy
 $(X_1)_n\text{RSEEGAP}(X_2)_m$, ahol X_1 és X_7 jelentése cisztein, és n és m jelentése egymástól függetlenül 0
 vagy 1.

12. A 11. igénypont szerinti peptid, ahol a peptid egy gyógyászati lag elfogadható hordozóhoz, előnyösen KLH-hoz (fűrócsiga hemocianin) kapcsolódik.

13. A 11. vagy 12. igénypont szerinti peptid, synucleinopátiák megelőzésében és/vagy kezelésében történő alkalmazásra.

14. Gyógyászati készítmény, amely legalább egy, a 11. vagy 12. igénypont szerinti peptidet tartalmaz.

15. A 14. igénypont szerinti gyógyászati készítmény, vakcinaként történő alkalmazás céljára.

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