COMPOSITION FOR TREATMENT OF MULTIPLE SCLEROSIS

FIELD OF INVENTION

The present invention relates to oligopeptides and fusion proteins and their use in the inactivation of epitope-specific anti MBP auto antibodies (ESAMBPCAA) involved into binding and catalytic degradation of MBP in course of progression of Multiple Sclerosis. The invention also relates to pharmaceutical compositions comprising said oligopeptides, combinations of the said oligopeptides and fusion proteins composed of the above-mentioned oligopeptides and their use for the treatment of multiple sclerosis.

BACKGROUND ART

Multiple sclerosis (hereinafter abbreviated as MS) is an inflammatory demyelinating disease of the human central nervous system with heterogeneous pathophysiological and clinical manifestations and a very complicated etiology (Hafler et al., 2004. J. Clin. Invest. 113: 788–794; Kornek et al., 2003. Brain Res. Bull. 61: 321–326.). Progression of the disease in humans leads to destruction of the myelin sheath and that ultimately affects the ability of nerves to conduct electrical impulses (Schwartz, R. S., 1993, in Fundamental Immunology, ed. Paul, W. E. (Raven, New York), pp. 1033–1097).

The viral mimicry hypothesis was formulated to explain the initiation of this pathology (Merkler et al., 2006. J. Clin. Invest. 116: 1254–1263.). At present, however, the true triggering mechanisms of the disease have not been clearly identified (Hohlfeld et al., 2004.Proc. Natl. Acad. Sci. USA 101 (Suppl. 2): 14599–14606).

It has been demonstrated that some of the proliferating T-cells in MS patients are directed towards MBP (Allegretta et al., Science, 247, 718-721, 1990) and that human T-cells can recognize multiple epitopes on the MBP molecule (Richert et al., J. Neuroimmun 23, 55-66, 1989). MBP also appears to be capable of activating some T-cells without the involvement of antigen presenting cells (Altman et al., Eur. J. Immun. 17, 1635-1640, 1987).

Despite strong and commonly accepted evidence for immune T cell involvement in development and progression of MS in humans and experimental animal models of the disease, the contributions of a specific B cell response to myelin sheath destruction is much less investigated (Klawiter et al.,2007. Curr. Neurol. Neurosci. Rep. 7: 231–238.; Nikbin et al., 2007. Int. Rev. Neurobiol. 79: 13–42.).

Ample data indicate that a significant portion of MS cases is characterized by the presence in the blood of auto antibodies directed against MBP components (Reindl et al., 1999, Brain, 122, 2047–2056; Genain et al., 1999, Nat. Med. 5,170–175). Moreover, high resolution microscopic analysis detected myelin-specific auto antibodies in the regions of demyelination plaques in human MS and a MS-like disease of marmosets, suggesting their direct contribution to myelin destruction (Genain et al., 1999, Nat. Med. 5,170–175). Although the mechanism of the autoantibody role in MS pathogenesis is unknown, auto antibodies to MBP and myelin oligodendrocyte glycoprotein (MOG) were proposed as biomarkers for clinical prognosis of MS (Berger et al., 2003, N. Engl. J. Med. 349, 139–145.10). Similar immunoglobulins were also found in mice with induced experimental allergic encephalomyelitis (EAE), which is an animal model of MS (Fritz et al., 1983, J. Immunol. 130, 191–194.). Increased titers of auto antibodies to MBP were observed in the cerebrospinal fluid (CSF) of patients with active forms of MS (Warren at al., Ann Neurol 209:20-25, 1986). Clinically, MS is characterized by phases of disease activity such as acute relapses or chronic progression, and by phases of clinical remission. Active MS is associated with increased levels of intrathecally appearing auto antibodies to MBP (Warren et al., Ann Neurol 209:20-25, 1986; Catz et al., Can J Neurol Sci 13:21-24, 1986). These antibodies are found predominantly in free (F) form during acute relapses and predominantly in bound (B) form when the disease is insidiously progressive (Warren et al., Ann Neurol 209:20-25, 1986). The therapy of relapsing-remitting multiple sclerosis (RRMS) patients with rituximab a monoclonal antibody, which selectively targets and depletes CD20+ B lymphocytes, appears safe and effective to some extend (Stüve, O. et al., Long-term B-lymphocyte depletion with rituximab in patients with relapsing-remitting multiple sclerosis., Arch Neurol. 2009 Feb;66(2):259-61).

Accordingly to that existing scientific background, the approach to treat MS using the immunomodulation with the whole MBP molecule and numerous peptides representing fragments and homologs of MBP protein and its fragments has been developed during last decade. As it is well known human MBP consists of 170 amino acid residues. In scientific and patent literature a fragment of MBP is labeled according to positions of its first and last amino acid residues counted from the C-terminus of full MBP sequence (SEQ ID NO: 1).

In WO 9612737 a number of peptides (human MBP fragments) having "T-cell activity" (i.e. the ability to influence on activity or functions of immune T-cells or their

subpopulations) were provided for preventing and treating of MS and has been disclosed as suitable for therapeutic use. To reveal the T-cell recognized epitopes in MBP, 16 short (each about 20 amino acid residues in length) and three more long (82-100, 83-105, 141-165) "overlapping" each other peptide sequences have been tested on their ability to stimulate of *in vitro* proliferation of peripherial blood cells of MS patients. The amino acid structure of peptides provided by inventors corresponds to the following MBP fragments: 11-30; 11-29; 11-31; 83-105; 82-105; 82-104; 80-98; 82-102; 80-104; 80-102; 111-130; 111-129; 141-165; 101-125. Also, these authors disclosure combination of the inventive peptides with others peptides known from prior art as having T-cell activity of MBP: 13 – 25; 31-50; 61-80; 82-92; 82-96; 82-97; 82-98; 82-100; 82-100; 83-100; 83-101; 84-97; 84-100; 85-100; 86-105; 87-99; 87-99 [91K>A]; 88-100; 88-99; 82-100; 111-135; 122-140; 139-170; 141-160; 142-166; 142-168; 146-160; 153-170.

Wraith D.C. and co-authors provide a method for selecting of tolerogenic peptides capable of binding to an MHC class I or II molecules without further antigen processing and using these peptides in a pharmaceutical composition for treatment and/or prevention of multiple sclerosis. A number of peptides corresponded to fragments 1-24; 15-39; 30-54; 45-69; 60-84; 75-99; 90-114; 105-129; 120-144; 135-159; 150 -170; 131-145; 132-146; 133-147; 134-148; 135-149; 136-150; 137-151; 138-152; 139-153; 140-154; 141-155; 142-156; 143-157; 144-158 of human MBP have been synthesized and used by these authors for identification of human MBP epitopes recognized by MS-patients' peripherial blood cells and then for study of ability of these peptides to bind to MHC class I or class II molecules. A number of short peptides , active in respect of modulation of immune T-cell functions were selected and claimed for MS treatment: 134-148; 135-149; 136-150; 137-151; 138-152; 139-153; 140-154; 30-44; 80-94; 83-99; 81-95; 82-96; 83-97; 84-98; 110-124; 130-144; 131-145; 132-146; 133-147 (see Applications: EP1918298, US 11/979,224, WO 03/64464).

In US2005209156 a peptide selected from MBP fragment (75-98) having the amino acid sequence of AGAPVVHPPLAIVTPAT including substitutions, additions or deletions thereof, has been disclosed for MS treatment.

In US Patent No. 5858980 the MBP fragments 84-102 and 143-168 were identified as containing immunodominant T-cell recognized epitopes of MBP active in the development of MS, and peptide comprising the amino acid sequence of ENPVVHFFKNIVTPRT (MBP fragment 83-98) and their analogs and

AQGTLSKIFKLGGRD (MBP fragment 146-160) as well as a pharmaceutical composition comprising the peptides were provided.

Warren K.G. with coauthors provides soluble synthetic peptides, useful to neutralize anti MBP auto antibodies, having the amino acid sequence correspond to following amino acid residues of human MBP: 61-75; 64-78; 69-83; 75-95; 69-83; 80-97; 91-106; 84-93; 85-94; 86-95; 87-96; 82-98. These peptides overlap MBP sequence from 61 to 106 of its amino acid residues (see WO 98/45327). One of the selected peptides (namely MBP 75-95) showed its ability to bound free antibodies to MBP both in vitro and in vivo (in blood of MS-patients). On the contrary, non-binding, control peptide MBP 35-58 had no effect on free and bound antibodies to MBP in MS patients.

Some of MBP based peptides and DNA vaccines has been evaluated in clinical trials : Bayhill Therapeutics DNA-vaccine BHT-3009 containing the gene for whole MBP sequence; altered peptide ligands GP77116 and NBI-5788 from Neurocrine Biosciences; oral tolerogenic composition Myloral from Autoimmune(R) based on whole MBP sequence. However no significant success was observed from those therapies due to serious adverse events or inability to slow MS progression (Hohlfeld et al., Proc Nat Acad. Sci U S A. 2004, October 5; 101(Suppl. 2): 14599–14606.). Now the abovementioned synthetic peptide with a sequence corresponding to amino acid residues 82-98 of MBP (DENPVVHFFKNIVTPRT) and coded as MBP8298 is being extensively studied in phase II of clinical trials for treatment of progressive multiple sclerosis. The peptide at dose of 500 mg was administered intravenously every six months. It has been shown that responders on the treatment with MBP8298 are the patients with HLA heliotypes DR2 and/or DR4 only (Warren, K.G. et al, European Journal of Immunology, 2006, vol. 13, No 8, pp. 887-895).

Above-mentioned MBP 84-102 fragment (US Patent No. 5858980) was used as active ingredient for preparation for treatment of secondary progressive MS in phase I of clinical trials. No treatment efficacy were detected according to measurement of Expanded Disability Status Scale, Nine Hole Peg Test, new lesions diagnosed by Magnetic Resonance Imaging (Goodkin, D. E. et al. Neurology 2000, vol. 54, pp. 1414-1420)

Thus the search and selection of novel fragments of MBP, oligopeptides having therapeutic activity against MS represents non-trivial task and medicinal properties of any of such oligopeptides with respect to MS treatment cannot be predicted beforehand by a

person skilled in the art even on the basis of extensive preclinical *in vitro* and/or *in vivo* testing. The identification and selection of such new effective peptides may be achieved through discovery of novel immunopathological mechanisms involved into development and progression of MS.

Earlier we have presented evidence that a small fraction of anti-MBP auto antibodies circulating in blood of MS patients demonstrates site-specific proteolytic cleavage of the MBP molecule and represents novel immunopathological mechanism of myelin destruction and progression of MS (Ponomarenko et al. Immunology Letters 103, 2006, pp. 45–50)

Accordingly, the object of the present invention is to determine novel oligopeptides which would be able to neutralize epitope-specific anti MBP auto antibodies involved into binding and catalytic degradation of MBP in course of progression of Multiple Sclerosis (ESAMBPCAA-epitope specific anti MBP catalytic auto antibodies), and new effective methods of treating MS using these novel oligopeptides.

This object is achieved by providing novel oligopeptide moieties (oligopeptides, fusion proteins composed of these oligopeptides and fragments of these oligopeptides), a pharmaceutical composition containing the said oligopeptides and fusion proteins, which neutralize ESAMBPCAA. Surprisingly, despite ESAMBPCAA represent only minor fraction of circulating anti MBP auto antibodies in MS, the inventors of the present invention have discovered that oligopeptides having sequences comprising as short as 6 amino acid residues of SEQ ID NO: 2, which neutralize ESAMBPCAA, demonstrate unexpectedly high efficacy in treatment of MS.

Other advantages and aspects of the present invention will become apparent upon reading the following detailed description of the invention.

SUMMARY OF THE INVENTION

In one embodiment, the present invention provides an oligopeptide (I) having amino acid sequence GGDRGAPKRGSGKDSHH (SEQ ID NO: 2).

In a further embodiment, the present invention provides an oligopeptide comprising one or more amino acid sequence alterations in SEQ ID NO: 2, wherein said oligopeptide contains at least 6 contigious amino acid residues of SEQ ID NO: 2 and is capable of binding ESAMBPCAA.

In still further embodiment, the present invention provides a fragment of oligopeptide I or a fragment of oligopeptide I comprising one or more amino acid sequence alterations in SEQ ID NO: 2, wherein said fragment has at least 6 amino acid residues in length and is capable of binding ESAMBPCAA.

In a further embodiment, the present invention provides a pharmaceutical composition comprising as an active ingredient a therapeutically effective amount of any one of the following oligopeptides:

- an oligopeptide I having amino acid sequence GGDRGAPKRGSGKDSHH (SEQ ID NO: 2);
- an oligopeptide comprising one or more amino acid sequence alterations in SEQ ID NO: 2, wherein said oligopeptide contains at least 6 contigious amino acid residues of SEQ ID NO: 2 and is capable of binding ESAMBPCAA
- a fragment of the oligopeptide I having amino acid SEQ ID NO: 2 or a fragment of an oligopeptide comprising one or more amino acid sequence alterations in SEQ ID NO: 2, wherein said fragment has at least 6 amino acid residues in length and is capable of binding ESAMBPCAA,

and a pharmaceutically acceptable carrier or diluent or drug delivery system.

In another embodiment, the present invention provides a pharmaceutical composition comprising as an active ingredient, a therapeutically effective amount of oligopeptide I having amino acid sequence SEQ ID NO: 2 or a fragment thereof, or an oligopeptide comprising one or more amino acid sequence alterations in SEQ ID NO: 2, or a fragment thereof, as set forth above and further comprising at least one oligopeptide selected from oligopeptide II having sequence SEQ ID NO: 3 and oligopeptide III having SEQ ID NO: 4.

In another embodiment, the present invention provides a fusion protein composed of one or more peptides having a sequence selected from the group of SEQ ID NO: 2, SEQ ID NO: 3, and SEQ ID NO: 4 or fragments thereof, which are linked sequentially to each other via a peptide or non-peptide linker in any order, wherein the fusion protein comprises at least one oligopeptide of SEQ ID NO: 2 or a fragment thereof.

In a preferred embodiment, a fusion peptide according to the present invention is composed of fragments of a sequence ID SEQ NO: 2, the fragments having at least 6 amino acid residues in length and linked sequentially to each other via a peptide or non-peptide linker.

In a further embodiment of the invention, a method for treating multiple sclerosis is provided, the method comprising administering an effective dose of an oligopeptide, or a fragment thereof, or a fusion peptide, or a pharmaceutical composition of the present invention to a subject in need thereof.

In a particular embodiment of the invention, the method for treating multiple sclerosis comprises: exposing the blood of a patient suffering from multiple sclerosis to an effective dose of an oligopeptide, or a fragment, or a fusion peptide or a pharmaceutical composition according to the present invention.

In another embodiment, use of an oligopeptide or a fusion protein of the present invention for manufacturing of a medicament for treating multiple sclerosis is provided.

BRIEF DESCRIPTION OF THE DRAWINGS

Figure 1 shows the amino acid sequence of intact human-MBP molecule. The block capitals represent amino acid residues on human MBP. Also the parts of MBP - amino acid sequence which correspond to amino acid sequence of inventive oligopeptide (SEQ ID NO: 2) as well as of oligopteptides having amino acid sequence SEQ ID NO: 3 and SEQ ID NO: 4, are presented correspondingly. These parts of MBP -amino acid sequence are outlined with black line.

Figure 2 shows degradation of MBP in presence of ESAMBPCAA and inhibition of degradation in presence of different MBP peptide fragments (the horizontal row of figures correspond to positions of first and last amino acid residues counted from the C-terminus of full MBP sequence).

Figure 3 illustrates the inhibition of ESAMBPCAA activity by different synthetic oligopeptides containing amino acid sequences of MBP fragment 43-68. Horizontal axis- concentration of oligopeptides used in inhibitory assay; Vertical axis- relative ESAMBPCAA activity (ESAMBPCAA activity in control assays (PBS) were deemed as 1). Codes of oligopeptides are indicated in Example 3.

Figure 4 illustrates suppression of MDS score of DA rats suffering from Experimental Allergic Encephalomyelitis and treated by inventive oligopeptides and the fusion protein. Suppression of MDS score in ongoing EAE in DA rats at day 24. Maximum clinical score in each group of rats, median and 95% confidential interval.

Figure 5 illustrates suppression of MDS score in ongoing experimental MS (Theiler's Virus Infection) with combinations of oligopeptides.

DETAILED DESCRIPTION OF THE INVENYION

DEFINITIONS

As used herein, the term "oligopeptide" relates to any molecule that contains up to about 20 amino acid residues linked by peptide linkage(s), i.e. term "oligopeptide" means a polypeptide up to 20 amino acids in length. The term "oligopeptide" or "oligopeptides" encompasses oligopeptides as well as salts thereof. Suitable salts include sodium or potassium salts or acetic or phosphate salts.

As used in this description, the term "oligopeptide" encompasses an "oligopeptide" having specified amino acid sequence and functional variants thereof. The functional variants include any fragment of the oligopeptide of specified sequence wherein the fragment has from 6 to 20 aminoacids in length provided that is retains capability of binding ESAMBPCAA.

As used in this description, the term an oligopeptide "fragment" of the present invention is comprised of at least about 6, preferably at least about 8, preferably at least about 12, more preferably at least about 14, and most preferably at least about 16 or more contiguous amino acid residues.

Further contemplated are variants of the oligopeptides of present invention obtained by altering the amino acid sequence of parent oligopeptide. Such alterations can comprise one or more amino acid substitutions, deletions, insertions or additions provided that resulting variant comprised at least 6 contigious aminoacids of the parent sequence and is capable of binding ESAMBPCAA.

As used in this description, the term "fusion protein" refers to an oligopeptide or a fragment thereof fused to another oligopeptide or a fragment thereof, each fused moiety being bound to the other directly by a peptide link (the amino group (NH2) of N terminus of one oligopeptide linked to the carboxylic acid group (COOH) of C-terminus of the other oligopeptide or a fragment thereof) or alternatively via a linker. The “linker” may be selected from “peptide linker” or “non-peptide linker”. A "peptide linker" may consist of a sequence containing from about 1 to about 20 amino acids, which are linearly linked to each other by peptide bond and which may optionally include a sequence for a protease cleavage site. The term “non-peptide linker” as used herein refers to any linking moiety having two or more reactive groups other than peptide linker. Preferred linker is a non-peptide polymer. The non-peptide polymer used a linker of the invention is a polymer carrying reactive groups at both ends, which are capable of independently binding to

reactive groups of an oligopeptide, wherein examples of reactive group of the oligopeptide includes a terminal amino group or a terminal carboxyl group, a lysine residue, a histidine residue or a cysteine residue. Reactive groups of the polymer include an aldehyde group, a propionic aldehyde group, a butyl aldehyde group, a maleimide group, a ketone group, a vinyl sulfone group, a thiol group, a hydrazide group, a carbonyldimidazole (CDI) group, a nitrophenyl carbonate (NPC) group, a trysylate group, an isocyanate group, and succinimide derivatives. Examples of succinimide derivatives include succinimidyl propionate (SPA), succinimidyl butanoic acid (SBA), succinimidyl carboxymethylate (SCM), succinimidyl succinamide (SSA), succinimidyl succinate (SS), succinimidyl carbonate, and N-hydroxy succinimide (NHS). The reactive groups at the ends of a non-peptide polymer may be the same or different. For example, the polymer may have a maleimide group at one end and an aldehyde group at another end. Low molecular weight linkers include carbodiimide or glutaraldehyde.

As used in this description, the term "ESAMBPCAA" refers to small fraction of autologous immunoglobulin molecules circulating in blood of MS patient which binds myelin basic protein and catalyze site-specific proteolytic cleavage of the MBP molecule.

As used in this description the terms "to treat," "treating," and "treatment" refer to administering a therapy, an agent, a compound, or composition to a subject suffering from a disease in order to reduce, ameliorate or to eliminate at least one symptom of the condition being treated as assessed by attending physician or any skilled in the art by any conventional method. As used in this description, the term 'Neutralization, inactivation, inhibition' means reduction of specific activity as measured with specified appropriate test system.

As used in the description, "effective amount" is an amount of an oligopeptide, a fragment thereof, or a fusion protein as described above, which upon administration, is capable of reducing ameliorating or eliminating at least one symptom of multiple sclerosis. Further, effective amount means an amount capable of reducing or preventing multiple sclerosis condition.

Based on our previous work (Belogurov at al., The Journal of Immunology, 2008, 180: 1258–1267) the novel class of auto antibodies involved into recognition and degradation of Myelin Basic Protein in MS patients was discovered. These catalytically active antibodies represent very minor fraction of anti MBP antibodies, however it was found that their catalytic activity correlates with progressive state of MS. Thus,

quantification of anti MBP autoantibody mediated MBP proteolysis was suggested as novel biomarker of MS. Surprisingly, despite ESAMBPCAA represent only minor fraction of circulating anti MBP auto antibodies in MS, the authors of present invention has discovered that certain oligopeptide , having strong neutralizing activity on ESAMBPCAA, demonstrate unexpectedly high efficacy in treatment of MS both in humans and in well-established animal models of multiple sclerosis. According to the present invention there an oligopeptide having amino acid sequence GGDRGAPKRGSGKDSHH (SEQ ID NO: 2) is provided. This sequence corresponds to amino acid sequence 46-62 of human MBP and is referred hereinafter as "MBP 46-62". Said oligopeptide is capable of inhibiting ESAMBPCAA.

Fragments and functional variants of oligopeptide GGDRGAPKRGSGKDSHH constitute one of preferred embodiments of present invention. On the basis of the ESAMBPCAA inhibition assays using a series of truncated forms of oligopeptide GGDRGAPKRGSGKDSHH the smallest number of amino acids required for oligopeptide fragment to retain its biological activity were established. Thus, according to the present invention, the fragment or functional variant of an oligopeptide GGDRGAPKRGSGKDSHH must retain at least 6 contigious amino acid residues of SEQ ID NO: 2. Based on given disclosure, it would be readily apparent to persons skilled in the art to determine, empirically, what variation can be made to the oligopeptide sequence GGDRGAPKRGSGKDSHH without affecting the ESAMBPCAA inhibitory activity of the peptide.

In another aspect, the present invention provides two oligopeptides having sequences GFGYGGRASDYKSAHK (SEQ ID NO: 3) and QGTLSKIFKLGGRDSRSGSPMARR (SEQ ID NO 4), respectively. These peptides contains well known in the art MBP epitopes, recognized by immune T – cells and/or by "conventional" (non-proteolytic) anti MBP auto antibodies from MS patients. The inventors have unexpectedly found that these two oligopeptides (SEQ ID NO: 3 and SEQ ID NO: 4 synergistically enhance efficacy of the oligopeptide (SEQ ID NO: 2) of present invention in treatment of multiple sclerosis in humans and preventing (or reducing) development of established animal model of MS- experimental allergic encephalomyelitis (EAE). Despite having low inhibitory activity against ESAMBPCAA as compared to oligopeptide (SEQ ID NO: 2), these two oligopeptides potentiate therapeutic effect of

GGDRGAPKRGSGKDSHH oligopeptide when co-administered to animals and humans. While not wishing to be bound by any theory, it may be assumed that this is due to fact that simultaneous modulation of several immune mechanisms - immune T cell function and auto antibody mediated MBP hydrolysis may provide multiplication of therapeutic effect. Accordingly, a pharmaceutical composition is provided which contains the oligopeptide of SEQ ID NO: 2, fragments or functional variants thereof and at least one of two oligopeptides of sequences SEQ ID NO: 3 or SEQ ID NO: 4.

In a further embodiment of the present invention, the oligopeptides of SEQ ID NO: 2, fragments or functional variants thereof are linked to each other to form a fusion peptide. Such fusion peptide comprises multiple copies of ESAMBPCAA inactivating moiety.

In a further embodiment of the present invention oligopeptide (SEQ ID NO: 2) or fragments or functional analogs thereof and oligopeptide (SEQ ID NO: 3) or fragments thereof and oligopeptide (SEQ ID NO: 4) or fragments thereof are linked to linked to each other in any order to form the fusion peptide, Thus, a fusion peptide of present invention may contain multiple repetitive copies of ESAMBPCAA inactivating moieties of oligopeptide (SEQ ID NO: 2) and epitopes of oligopeptides (SEQ ID NO: 3) and (SEQ ID NO: 4).

The invention further provides a pharmaceutical composition comprising therapeutically effective amount of an oligopeptide of present invention (SEQ ID NO: 2) or fragment or functional variant thereof or fusion protein and a pharmaceutically acceptable carrier or a diluent and / or drug delivery system. Examples of pharmaceutical acceptable carriers are well known in the art, and include for example normal saline. The examples of drug delivery systems are also well known in the art and include for example liposomes or synthetic polymeric nanoparticles. The oligopeptides of the present invention can be prepared according to existing methods of synthesizing oligopeptides having formula according disclosure provided. Fusion proteins may be produced by recombinant DNA technology. Knowing the sequence of the selected fusion proteins, as disclosed in the present invention, the appropriate DNA sequence can be produced by conventional, known methods of synthesizing DNA sequences. The DNA sequences so produced can then be cloned into appropriate cloning vehicles and used to transform an appropriate host cell to produce the recombinant peptide. All of the methodology referred to above is conventional and well-known to persons skilled in the art.

The invention further provides a method for treating multiple sclerosis, the method comprising administering an effective dose of oligopeptide, or a fragment, or fusion peptide, or a pharmaceutical composition containing same to a subject in need thereof.. The therapeutic dose of oligopeptides or fusion protein for the treatment of MS may be from about 0,01 mg per kilogram of body weight to about 10.0 mg per kilogram of body weight; the composition can be administered intravenously, subcutaneously, intrathecally. In one example of the present invention, the composition is administered orally, to target so called "mucosal delivery route". The composition can be administered as a single or sequential dose, as may be required.

While this invention is described in detail with particular reference to preferred embodiments thereof, the following examples are offered to illustrate but not limit the invention.

EXAMPLES

Example I. Detection of ESAMBPCAA -autologous immunoglobulin molecules which binds myelin basic protein and catalyze site-specific proteolytic cleavage of the MBP molecule in blood of Multiple Sclerosis Patients

Anti MBP autoantibody purification and characterization were done using the serum of 24 MS patients (17–54-year-old, mean age 32 years) who had not been treated with steroids or non-steroidal anti-inflammatory drugs. The MS diagnosis was verified and confirmed, and EDSS (Expanded Disability Status Scale) values were calculated according to Poser's classification of disease progression, using clinical, immunological and MRI data (Poser et al., Clin Neurol Neurosurg 2001; 103:1–11.). Immunoglobulins (IgG) were isolated from serum by thrice-repeated 50% ammonium sulfate precipitation followed by affinity chromatography on protein G-Sepharose (Amersham Biosciences). IgG-containing fractions were then dialyzed against PBS or TBS with 0.05% NaN_3 at 4°C. The IgG amount was quantified and standardized by ELISA. IgG purity was assessed by electrophoresis followed by silver staining, immunoblotting under nonreducing conditions and by surface-enhanced laser desorption/ionization (SELDI) mass spectrometry. IgGs were further separated by the antigen affinity chromatography on a column with MBP immobilized on NHS-Sepharose (Amersham) and their purity was assessed by electrophoresis followed by silver staining technique.

MBP was prepared from bovine brain according to Miller (Miller et al., 1996. Experimental autoimmune encephalomyelitis in the mouse. In: Current Protocols in Immunology; J. E. Coligan, ed. Wiley, New York, p. S19.) The resulting protein was purified by reverse phase HPLC on column C4 10/250 (Mashery-Nagel, Germany).

MBP hydrolysis by anti MBP auto antibodies was assayed as follows: Purified antibodies (0.1–1mkg) were incubated at 37 ◦C for 14 h in the final volume of 12.5mkl PBS, 0.02% NaN_3 containing 1–2 mkg of MBP. The samples were mixed with Laemmli's buffer. The extent of MBP degradation was visualized by SDS-PAGE in Tris–glycine and Tricine buffer systems. For quantitative MBP degradation assay, MBP (10 mkM) was incubated at 37 ◦C with antibodies (60 nM) in 0.1 ml of PBS, 0.02% NaN_3 for 12 h. The reaction was stopped by adding 10% TFA up to pH 2.5. The samples were further chromatographed on column C4 4.0/150 (Waters). The amount of non-cleaved MBP was calculated by absorbance monitoring at 280 nm.

The results are presented in Table 1.

Table 1: Clinical statuses of 24 multiple sclerosis patients and corresponding rates of MBP hydrolysis by ESAMBPCAA.

Patient N	Type of disease	ESAMBPCAA activity (pmol/min/nmol)	EDSS score
MS1	SP	77,6+7,1	5,0
MS2	SP	84,3+6,9	4,5
MS3	SP	63,0+5,8	6,0
MS4	RR	45,6+6,0	2,0
MS5	RR	5,9+4,8	1,0
MS6	RR	2,1+4,5	1,0
MS7	RR	5,3+5,0	2,0
MS8	RR	3,0+3,9	1,5
MS9	SP	71,2+5,5	3,0
MS10	SP	4,5+4,1	2,5
MS11	RR	1,5+6,0	0
MS12	SP	79,0+8,9	4,0

MS13	RR	2,0+4,0	0
MS14	PP	44,9+5,0	3,0
MS15	PP	22,3+4,5	3,0
MS16	RR	29,3+5,3	2,5
MS17	RR	15,1+4,1	3,0
MS18	RR	4,3+3,9	1,5
MS19	RR	6,5+4,0	2,0
MS20	SP	15,0+5,9	3,0
MS21	SP	1,3+3,7	2,0
MS22	RR	39,2+7,2	3,0
MS23	SP	29,1+6,2	3,0
MS24	RR	4,3+3,5	1,5

SP - secondary progressive; PP-primary progressive; RR- relapsing–remitting.

Importantly, the level of the ESAMBPCAA mediated MBP cleavage correlated with expanded disability status scale (EDSS) range of the patients ($r2 = 0.85$, $P < 0.001$ by Spearman rank correlation). The highest levels of the antibody mediated catalysis occurred in cases with high EDSS (from 3.0 to 6.0), mostly at the progression stage or the exacerbation at the relapsing–remitting (RR) course of the disease.

Example 2. Screening for ESAMBPCAA inhibitory sequences.

Twelve DNA fragments encoding human MBP peptides representing different parts of MBP molecule (1–27, 17–41, 25–54, 43–68, 53–81, 81–103, 91–114, 107–132, 123–140, 130–156, and 146–170, were prepared by PCR with four overlapping corresponding to the linker (SGGGG)3S. The final PCR products were cloned in-frame into pET32CH plasmid by using NcoI and BamHI restriction sites. The expression products of these plasmids that contained Trx fused to MBP peptides were used for cleavage analysis. The plasmid encoding Trx (Thioredoxin) with linker (SGGGG) 3S was designed for control. The soluble recombinant His-tagged proteins were obtained by *Escherichia coli* expression and isolated by sorption on Talon SuperFlow (BD Biosciences) column, followed by cation exchange chromatography on Mono S column (Amersham Pharmacia)

at pH 5.0 and subsequent size exclusion chromatography on Superdex 75 GL 10_300 column (Amersham Pharmacia) in 150 mM NH_4HCO_3 buffer.

Pattern of MBP hydrolysis by ESAMBPCAA purified from plasma of SJL mice suffering from EAE, induced by injection of MBP in complete Freund adjuvant, was assessed by SDS-PAGE as specified in Example 1 in presence of 0,5 mkM of different MBP peptides. The results are presented in Figure 2. The MBP fragment 43-68, which contains sequence GGDRGAPKRGSGKDSHH, demonstrates potent inhibitory activity on anti MBP auto antibody mediated MBP hydrolysis.

Example 3. Suppression of MBP-specific auto antibody mediated hydrolysis by synthetic oligopeptides containing amino acid sequences of MBP fragment 43-68.

In order to identify the amino acid sequence required for efficient inhibition of ESAMBPCAA activity the following series of peptides were synthesized:

Table 2:

	Amino acid sequence	Number of amino acid residues	Position in MBP sequence
A1	RFFGGDRGAPKRGSGKDSHHPARTAH	26	43-68
A2	FGGDRGAPKRGSGKDSHHPAR	21	45-65
A3	GGDRGAPKRGSGKDSHH	17	46-62
A4	GAPKRGSGKDSHH	13	50-62
A5	GGDRGAPKRGS	11	46-56
A6	PKRGSGKDSHH	11	52-62
A7	GGDRGAPKR	9	46-54
A8	RGSGKDSHH	9	54-62
A9	GGDRGAP	7	46-52
A10	SGKDSHH	7	56-62
A11	GGDRG	5	46-50
A12	KDSHH	5	58-62

The oligopeptides were assayed for their potency to inhibit ESAMBPCAA mediated MBP hydrolysis using ESAMBPCAA collected from progressive MS Patient as specified in Example 1. Different concentrations of test peptides were mixed with both antibodies (30 nM) and MBP (4 mkM) in TBS with_0.1% NaN3_and 10 mM CaCl2. The samples were incubated for 16 h at 37°C and analyzed on 15% SDS_PAGE. The gels were stained by Coomassie and analyzed by densitometry with TOTALLAB 2.01 software (Nonlinear Dynamics, Ltd., Newcastle upon Tyne, U.K.). The data presented in Figure 3 shows that from tested oligopeptides the oligopeptide A3, comprising amino acid sequence GGDRGAPKRGSGKDSHH demonstrates most potent inhibitory activity, having less amino acid residues than oligopeptides A1 and A2. Shortening of oligopeptide lengths to less than 6 amino acids lead to complete lack of ESAMBPCAA inhibitory activity.

Example 4. Suppression of MBP-specific auto antibody mediated hydrolysis by synthetic oligopeptides containing amino acid sequence GGDRGAPKRGSGKDSHH, fragments thereof and fusion proteins.

Serum samples collected from 5 patients suffering from progressive MS were quantified for ESAMBPCAA mediated MBP hydrolysis as specified in Example 1 in presence of following substances:

A1- negative control: phosphate buffered saline (PBS);

A2 – glatiramer acetate (Copaxone, Teva Pharmaceuticals), 100 nM;

A3- oligopeptide GGDRGAPKRGSGKDSHH 100 nM;

A4- oligopeptide APKRGSGKDSH (a fragment of oligopeptide GGDRGAPKRGSGKDSHH - 100nM;

A5-oligopeptide SGKDS (a fragment of oligopeptide GGDRGAPKRGSGKDSHH)-100 nM;

A6- fusion protein RGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGK (containing repetitive sequence RGAPKRGSGK, which sequence represents the fragment of GGDRGAPKRGSGKDSHH oligopeptide-100 nM;

The products A3, A4 and A5 have been synthesized by the laboratory of solid phase organic synthesis (Shemyakin Ovchinnikov Institute of Bioorganic Chemistry RAS. The

product A6 has been synthesized using ABNOVA custom cell-free translation system. (www.abnova.tw).

The results of the assay are presented in Table 3 below.

Table 3. Influence of different tested preparations on MBP-specific auto antibody mediated hydrolysis (pmol/min/nmol)

	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5
A1 (PBS)	77.6 ± 7.1	84.3 ± 6.9	63.0 ± 5.8	45.6± 6.0	79.0±8.9
A2	62,5±7,2	72,6±7,7	51,2±3,5	47,7±7,9	65,7±5,4
A3	11,2±3,5	17,3±4,1	14,4±2,2	14,5±4,4	16,9±7,3
A4	12,7±3,6	15,6±2,9	14,3±4,4	15,8±4,9	13,3±5,9
A5	57±4,1	59,7±5,7	44,2±6,6	45,5±3,8	55,7±6,1
A6	7,7+3,3	22,3±4,2	20,1±3,9	21,5±6,6	5,0±1,8

Thus, oligopeptide GGDRGAPKRGSGKDSHH, oligopeptide APKRGSGKDSH (a fragment of oligopeptide GGDRGAPKRGSGKDSHH with 11 amino acids length and the fusion protein containing repetitive sequence RGAPKRGSGK, which sequence represents the fragment of GGDRGAPKRGSGKDSHH oligopeptide all has potent suppressive effect on MBP-specific auto antibody mediated hydrolysis

Example 5

Treatment of EAE by the inventive oligopeptide and the fusion protein

Experimental Allergic Encephalomyelitis (EAE) in DA rats is well established animal model of MS. The animal work was performed in the Pushchino branch of the Shemyakin and Ovchinnikov Institute of Bioorganic Chemistry, Russian Academy of Sciences. Female DA rats 10–14 weeks of age were anesthetized and injected intradermally at the base of the tail with 100 ml of inoculums containing 50 mg of rat MOG in saline emulsified (1:1) with CFA (Sigma Chemical Co., St. Louis, MO) containing 200 mg of *Mycobacterium tuberculosis* (strain H 37 RA; Difco Laboratories, Detroit,MI). At day 8 after a second immunization, rats were divided into 6 groups (6 rats per group) and treated during 5 days (days 8-12) according following schedule:

Group 1- oligopeptide GGDRGAPKRGSGKDSHH at 120 mkg daily by intranasal administration

Group 1- oligopeptide GGDRGAPKRGSGKDSHH at 50 mkg daily by intranasal administration

Group 3- fusion protein RGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGK at 120 mkg daily by intranasal administration

Group 4- oligopeptide APKRGSGKDSH at 120 mkg daily by intranasal administration

Group 5- Glatiramer acetate (Copaxone, Teva) 120 mkg daily by intranasal application

Group 6- Control (PBS)

Clinical symptoms (MDS score) of disease were assessed daily on the following grading scale: grade 0, no clinical signs; grade 1, mild waddling gait or flaccid tail; grade 2, severe waddling gait; grade 3, moderate hind limb paresis; and grade 4, severe hind limb paralysis. Serum samples collected from animals were quantified for ESAMBPCAA mediated MBP hydrolysis as specified in Example 1

The results are presented at Figure 4 and Table 4 below.

Table 4. Influence of treatment on rates of MBP-specific auto antibody mediated hydrolysis (Median for 6 animals; pmol/min/nmol)

Experimental Group	Day 8	Day 16	Day 24
Group 1	42,8+5,5	17,3+5,4	15,1+3,9
Group 2	43,1+6,4	35,6+4,9	46,3+2,9
Group 3	45,7+6,5	48,1+7,4	39,4+4,6
Group 4	49,3+4,9	45,9+7,1	45,1+6,3
Group 5	51,4+8,2	55,4+5,7	62,2+5,7
Group 6	47,6+6,1	61,3+7,35	75,2+8,3

Thus, both the MBP-specific auto antibody mediated hydrolysis and MDS score are most efficiently suppressed by oligopeptide GGDRGAPKRGSGKDSHH in the dose-dependent manner. Fusion protein

RGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGA PKRGSGK and oligopeptide APKRGSGKDSH are also active in suppression of MBP-specific auto antibody mediated hydrolysis and MDS score.

Example 6

Treatment of experimental MS (Theiler's Virus Infection) with inventive oligopeptides and combinations of oligopeptides

The BeAn strain of TMEV used in this study was generated and propagated in BHK-21 cells grown in Dulbecco's modified Eagle's medium supplemented with 7.5% donor calf serum. For i.c. infection, 30 µl of virus solution (0.2 x 10^6 to 6 x 10^6 PFU) was injected into the right cerebral hemisphere of 6- to 8-week-old SJL mice (8 animals per group) anesthetized with isoflurane. Clinical symptoms (MDS score) of disease were assessed weekly on the following grading scale: grade 0, no clinical signs; grade 1, mild waddling gait or flaccid tail; grade 2, severe waddling gait; grade 3, moderate hind limb paresis; and grade 4, severe hind limb paralysis. Serum samples collected from animals were quantified for ESAMBPCAA mediated MBP hydrolysis as specified in Example 1

Oligopeptides GFGYGGRASDYKSAHK (SEQ ID NO: 3) and QGTLSKIFKLGGRDSRSGSPMARR (SEQ ID NO: 4) are known in the art as capable to modulate immune T cell activity in MS, leading to some efficacy in treatment of MS.

At day 14 following infection mice were divided into 6 groups (30 mice per group) and treated during 10 days (days 14-24) according following schedule:

Group 1- Control -PBS daily by subcutaneous injection

Group 2- Copaxone daily 30 mkg daily by subcutaneous injection

Group 3- Oligopeptide GGDRGAPKRGSGKDSHH at 20 mkg daily by subcutaneous injection

Group 4- Oligopeptide GFGYGGRASDYKSAHK at 20 mkg daily by subcutaneous injection

Group 5- Oligopeptide QGTLSKIFKLGGRDSRSGSPMARR at 20 mkg daily by subcutaneous injection

Group 6- Oligopeptide GGDRGAPKRGSGKDSHH at 20 mkg and oligopeptide GFGYGGRASDYKSAHK at 20 mkg daily by subcutaneous injection

Group 7- Oligopeptide GGDRGAPKRGSGKDSHH at 20 mkg and oligopeptide GFGYGGRASDYKSAHK at 20 mkg and oligopeptide QGTLSKIFKLGGRDSRSGSPMARR at 20 mkg daily by subcutaneous injection

Group 8 - Oligopeptide GFGYGGRASDYKSAHK at 20 mkg and oligopeptide QGTLSKIFKLGGRDSRSGSPMARR at 20 mkg daily by subcutaneous injection

The results are presented at Figure 5 and Table 5 below.

Table 5. Influence of performed treatment on disease manifestation and rates of auto antibody mediated MBP hydrolysis.

Group	% Animals affected with disease at day 100	ESAMBPCAA activity at day 100 (pmol/min/nmol)
Group 1	27 (30)	45,7+6,7
Group 2	25(30)	39,8+4,1
Group 3	19(30)	11,4+3,5
Group 4	25(30)	40+3,1
Group 5	27(30)	35,3+5,1
Group 6	15(30)	16,5+3,2
Group 7	17(30)	14,1+2,9
Group 8	25(30)	37,7+4,9

Thus, MBP-specific auto antibody mediated hydrolysis and MDS score are most efficiently suppressed by oligopeptide GGDRGAPKRGSGKDSHH alone or in combination with oligopeptide GFGYGGRASDYKSAHK and/or oligopeptide QGTLSKIFKLGGRDSRSGSPMARR.

Oligopeptides GFGYGGRASDYKSAHK and oligopeptide QGTLSKIFKLGGRDSRSGSPMARR alone or in combination has moderate therapeutic effect and do not affect MBP-specific auto antibody mediated hydrolysis. Combination of GGDRGAPKRGSGKDSHH with GFGYGGRASDYKSAHK and QGTLSKIFKLGGRDSRSGSPMARR has most beneficial therapeutic effect. Importantly, peptide GGDRGAPKRGSGKDSHH and its combinations with oligopeptide

GFGYGGRASDYKSAHK and/or oligopeptide QGTLSKIFKLGGRDSRSGSPMARR have also disease-preventive (prophylactic) activity.

Example 7

Suppression of MBP-specific auto antibody mediated hydrolysis and treatment of MS by means of exposing the blood of patient to the fusion proteins according the invention:

Three fusion proteins of the following formulas were synthesized using ABNOVA custom cell-free translation system (www.abnova.tw):

1)
RGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGKRGAPKRGSGK

2)
GAPKRGSGKYGGRASDYKSGTLSKIFKLGGRDSRRGAPKRGSGKYGGRASDYKSGTLSKIFKLGGRDSR

3)
GGDRGAPKRGSGKDSHHGFGYGGRASDYKSAHKQGTLSKIFKLGGRDSRSGSPMARR

Thus, these fusion proteins contain SEQ ID NO: 2, SEQ ID NO:3, SEQ ID NO:4 linked in a different order.

All three oligopeptides were coupled to CNBr-activated Sepharose beads (Pharmacia) at 0, 1 mg of each oligopeptide per 1 ml beads under aseptic conditions. After coupling, the oligopeptide-Sepharose was washed extensively with 0.01M Tris-buffered 1.14 M NaCl, pH 8.0 containing 10 mM EDTA to remove noncovalently associated material and packed into 400 ml columns.

The case of ESAMBPCAA apheresis was performed in St.Petersburg Medical Academy, Department of Neurology. We performed the ESAMBPCAA apheresis in 18-year old patient with rapidly progressing severe secondary progressive MS, deemed being resistant to conventional therapy (corticosteroids and immunosuppressants). The patient underwent 5 apheresis cycles, one cycle per week. Patient plasma was separated by a continuous-flow plasma separator, the Cobe Spectra, USA. The blood flow varied from 50 to

70 ml/min. Heparin was also added with an initial bolus of 5000 U, then, in the first phase, 50 U/min by the pump. Plasma was than washed through the Sepharose column, and the rest of blood was returned to the patient. The flow of plasma through the Sepharose column was controlled by a computerized adsorption–desorption device ADA—Medicap. Following passage through the column the plasma returned to the patient. In each cycle 3000 ml of plasma was treated. The EDSS score dropped from 6 before the first apheresis to 5.5 two weeks after the last cycle. The control MRI performed one month after the last cycle showed stabilization of the previous lesions with the reduction of two lesions. No new lesions were detected. The rate of MBP-specific auto antibody mediated hydrolysis was assayed as specified in Example 1 before and two weeks after the last apheresis cycle. The data is presented in the Table 6 below.

Table 6. Suppression of MBP-specific auto antibody mediated hydrolysis under the treatment performed.

Parameter	Before therapy	2 weeks following last cycle
ESAMBPCAA	87 pmol/min/nmol	15 pmol/min/nmol
IgG	410 mg/dL	395 mg/dL
IgA	53 mg/dL	57 mg/dL
IgM	24 mg/dL	22 mg/dL
EDSS score	6	5,5

Thus, exposing the blood of MS patient to fusion proteins according the invention suppresses ESAMBPCAA activity and provides effective treatment for MS.

Also, certain other fusion proteins containing SEQ ID NO: 2, SEQ ID NO:3, SEQ ID NO:4 linked in other order (via peptide and non-peptide linkers) were produced and successfully tested for ESAMBPCAA suppressing activity and ability to ameliorate MS symptoms and to decrease MS biomarker.

Example 7

Pharmaceutical composition containing fragments of oligopeptide

The three oligopeptides of following formulas: GGDRGAPKRGSGKDSHH, GFGYGGRASDYKSAHK and QGTLSKIFKLGGRDSRSGSPMARR have been synthesized by the laboratory of solid phase organic synthesis (Shemyakin Ovchinnikov Institute of Bioorganic Chemistry RAS); phospholipids were purchased from Sigma – Aldrich and Avanti Polar Lipids. 50g of mixture of egg phosphatidylcholine (PC),

Dioleoyl phosphatidylcholine (DOPE) and 1, 2-dioleoyl-3- trimethylammonium-propane (DOTAP) (at 4:2:1 molar ratio) was dissolved in chloroform and then chloroform was removed by evaporation. Oligopeptides 1, 2 and 3 were dissolved in phosphate buffered saline (PBS), PH7, 4 at 5 g/L concentration of each individual oligopeptide (15 g/L total protein). The lipid film in the evaporation vessel was rehydrated by mixing with 10 litres of PBS-peptides solution and 30 min shaking at 40°C.The resulting lipid/protein emulsion was transferred to 10 ml vials and lyophilized. The dry powder was solubilised "ex-tempore" with 5 ml of water for injection (WFI) to form large multi-lamellar liposomes (LMV) containing the oligopeptides.

Example 8

Treatment of patients with relapsing remitting MS with oral liposomal formulation of oligopeptides

The trial was performed between April 2003 and June 2005 in St.Petersburg Medical Academy, Department of Neurology. Under the approval of local Ethics Committee totally 15 patients(5 male and 10 female with median age of 32 years) with relapsing-remitting MS (diagnosed according Poster and MacDonald criteria) with at least two documented relapses within two last years and having EDSS (Kurtzke Expanded Disability Status Scale) score from 0 up to 5,5 were recruited to the trial.

Patients were treated by daily oral dosing of liposomes (LMV), manufactured as described in Example 8 at 0, 5 mg of protein per kg of body weight. The treatment was performed as monotherapy continuously daily for the period of 2 years.

EDSS scoring were checked once every two month. Brain MRT check for active demyelinization locuses (ADL) was performed once every two month. Analysis of frequency and duration of relapses was performed at the end of 2-year observation period. Blood cell counts, ESAMBPCAA activity, blood biochemistry and immunological parameters were checked at the beginning and the end of the trial.

All 15 patients have stable EDSS score within first 6 month of treatment. The average frequency of relapses decreased from 2, 7 per year at inclusion to 1, 5 per year at the end of treatment period. During 2 year follow-up period 11 patients were relapse-free. 6 patients (40%) have less ADL at the end of treatment period. 7 patients (47%) have process stabilization -the same number of ADL. In 2 patients (13%) the ADL count has increased. No adverse events linked to investigational preparation were noted. The

immunological parameters, blood biochemistry and blood cell counts were not significantly changed at the end of treatment period. The data is presented in Table 7.

Table 7. Immunological, blood and blood biochemistry parameters at the beginning and at the end of treatment (data average for 15 patients)

Parameter	Before treatment	After treatment
CD3	48,75+9,68	52,37+7,37
CD4	29,75+6,43	30,25+4,06
CD8	22,87+5,34	25,5+6,2
CD4/CD8	1,37+0,29	1,29+0,35
IgA	2,88+1,53	2,77+1,46
IgM	1,59+0,71	1,68+0,41
IgG	19,67+6,45	16,00+5,10
HLADR	23,13+5,38	21,25+5,88
ESAMBPCAA	45,1+6,2	12,5+4,5
HB	128,86+14,73	126,57+20,08
WBC	5,69+1,38	5,04+0,98
Lymphocytes %	28,29+8,94	25,36+5,12
ALT	0,29+0,20	0,21+0,07
AST	0,25+0,12	0,19+0,06
Bilirubin	12,36+4,17	10,39+2,67
Urea	3,62+1,67	4,11+1,16
Creatinine	0,07+0,02	0,08+0,01

Lower rate of MBP-specific auto antibody mediated hydrolysis was noted and that was in coincidence with significant curative effect achieved. Thus, the oral intake of oligopeptides according to present invention has favorable therapeutic performance associated with decrease of ESAMBPCAA activity. Under comparison with retrospective data available, results of treatment according the present invention shows better results than those of Beta-Interferon, which is existing standard of MS therapy. The comparison table 8 is presented below.

Table 8.

Comparative data on clinical efficacy of currently available standard treatment regimes of MS and results of treatment according Example 8.

Treatment	Clinical Observations	MRI Data: decrease of active lesions	Adverse Reactions	Reference
Copolymer 1	Decrease of relapse frequency - 29%; Slowing EDSS score progression	No statistical significant decrease	Low	Johnson KP, Brooks BR, Cohen JA, Ford CC, Goldstein J, Lisak PP et al. Copolymer-1 reduces relapse rate and improves disability in relapsing-remitting multiple sclerosis: Results of a phase III multicentre, double-blind, placebo controlled trial. The copolymer-1 multiple sclerosis study group. Neurology 1995; 45:1268.
Beta-Interferon	Decrease of relapse frequency - 83%; No slowing EDSS score progression	83% decrease	High	Paty DW, Li DKB, the UBC MS/MRI Study Group and the Interferon Beta Multiple Sclerosis Study Group. Interferon beta-1b is effective in relapsing-remitting multiple sclerosis. II. MRI results of a multi-centred, double-blind, placebo-controlled trial. Neurology 1993;43:662-7
Oligopeptides according the invention	Decrease of relapse frequency -87%; Slowing EDSS score progressior	75% decrease	No	Example 8

The foregoing description of the present invention provides illustration and description, but is not intended to be exhaustive or to limit the invention to the precise one disclosed. Modifications and variations are possible consistent with the above teachings or may be acquired from practice of the invention. Thus, it is noted that the scope of the invention is defined by the claims and their equivalents.

CLAIMS

1. An oligopeptide having amino acid sequence GGDRGAPKRGSGKDSHH (SEQ ID NO: 2).
2. An oligopeptide of claim 1 comprising one or more amino acid sequence alterations in SEQ ID NO: 2, wherein said oligopeptide contains at least 6 contigious amino acid residues of SEQ ID NO: 2 and is capable of binding anti Myelin Basic Protein hydrolyzing auto antibodies (ESAMBPCAA).
3. A fragment of oligopeptide of any of claims 1 or 2, wherein said fragment has at least 6 amino acid residues in length and is capable of binding anti Myelin Basic Protein hydrolyzing auto antibodies (ESAMBPCAA)
4. A pharmaceutical composition comprising therapeutically effective amount of an oligopeptide of any one of claims 1 or 2, or a fragment according to claim 3 as an active ingredient and a pharmaceutically acceptable carrier, excipient or drug delivery system.
5. A pharmaceutical composition of claim 3 wherein said pharmaceutical composition further contains at least one oligopeptide selected from oligopeptides having sequences SEQ ID NO: 3 and SEQ ID NO: 4.
6. A fusion peptide composed of fragments of SEQ ID NO: 2, having at least 6 amino acid residues in length, and linked sequentially to each other via peptide or non-peptide linker.
7. A fusion peptide composed of one or more peptides having sequence selected from the group including SEQ ID NO: 2, SEQ ID NO: 3 and SEQ ID NO: 4, or fragments thereof, which are linked sequentially to each other via peptide or non-peptide linker in any order, wherein the said fusion peptide comprises at least one oligopeptide having amino acid sequence SEQ ID NO: 2 or a fragment thereof.
8. A pharmaceutical composition comprising a therapeutically effective amount of a fusion protein of any one of claims 6 or 7 as an active ingredient and a pharmaceutically acceptable carrier, excipient or drug delivery system.
9. A method of treating or preventing multiple sclerosis in a subject in need of such treatment, wherein the said method comprises administering to the subject a composition according to any one of claims 4, 5 or 8.
10. The use of a composition according to any one of claims 4, 5 or 8 in the manufacture of a medicament for treating multiple sclerosis.