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(54) Title: MULTISPECIFIC BINDING MOLECULES FOR THE PREVENTION, TREATMENT AND DIAGNOSIS OF NEURODEGENERATIVE DISORDERS

(57) Abstract: The invention relates to a multispecific binding molecule, preferably an antibody, comprising at least a first binding site binding to a first target selected from the group comprising human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidized, aggregated, cleaved human tau protein and truncated versions thereof, α -synuclein, TDP43, mHTT, and fragments thereof and at least a second binding site binding to a second target selected from the group comprising human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidized and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, TDP43, mHTT, C1q, C5a, IL1B, IL6, TNF- α , APOE, IL12, IL23 and fragments thereof, wherein the first and second target; are different. Further, the invention relates to a method, a diagnostic method and use of the binding molecules, specifically antibodies of the invention for the treatment and prevention of neurodegenerative diseases.



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Title: Multispecific binding molecules for the prevention, treatment and diagnosis of neurodegenerative disorders

5 FIELD OF THE INVENTION

The present invention relates to multispecific binding molecules, as well as use of these binding molecules in the diagnosis or treatment of neurodegenerative disorders, such as Alzheimer's Disease, Lewy Body Dementia and Parkinson's Disease.

10 BACKGROUND OF THE INVENTION

Neurodegenerative disorders (NDDs) are a group of conditions that is characterized by the progressive loss of structure and function of the central and peripheral nervous system. Examples of neurodegenerative disorders include Alzheimer's Disease (AD), Parkinson's Disease (PD), Huntington's Disease (HD) and Amyotrophic Lateral Sclerosis (ALS). Most NDDs are associated with the aggregation and deposition of misfolded proteins, causing toxicity to the directly affected and surrounding cells, which leads to the dysfunction and loss of synaptic connections, neuroinflammation and can ultimately result in the death of neurons. These processes lead to cognitive dysfunction and memory impairment, as well as motor dysfunction and specific clinical symptoms depending on the area in the brain where the aggregates are located.

Aggregation of misfolded proteins, resulting in the formation of insoluble filaments in the brain, is a characteristic process in neurodegeneration. Although misfolding of proteins frequently occurs in healthy subjects as well, these abnormal and aggregated proteins are usually efficiently cleared or repaired by the system. However, in diseased subjects these clearing mechanisms are impaired or overwhelmed, hence continued aggregation and ultimately deposition of these aggregates occurs. The soluble oligomers and short fibrils are thought to be the most toxic as they are highly reactive molecules that can diffuse throughout the cell and abnormally interact with cellular proteins. Furthermore, intracellular aggregated proteins can be secreted into extracellular space (e.g. via synaptic release or membrane vesicles) and are subsequently taken up by neighboring or synaptically connected neurons. These protein aggregates incorporate their physiological monomeric proteins in these new cells. This process is called 'seeding' and leads to a vicious cycle of protein aggregation and subsequent excretion to other cells. This process leads to the so-called propagation of protein pathology throughout the brain and is thought to contribute to the progressive nature of the protein pathology in many NDDs. There is also substantial evidence that aggregated proteins can induce misfolding and subsequent aggregation of other proteins due to direct molecular interactions, a phenomenon known as "cross-seeding". In addition, the presence of protein aggregates can possibly lead to impairment of the clearance system of the cell and therefore other aggregates comprising different proteins are also often observed. Furthermore, since age is the biggest risk factor for most sporadic (non-familial) NDDs, it would be predicted by chance that elderly people have diverse pathology in their brain. Hence, in many NDDs protein aggregates of multiple different protein types have been observed. Disorders that are associated with the accumulation of abnormal proteins are also frequently called "proteopathies". Proteins commonly observed to form aggregates in NDDs are

tau-protein, amyloid- β , α -synuclein, TDP-43 and mutant Huntingtin protein (mHTT). (Skovronsky, D.M., et al., *Annu. Rev. Pathol. Mech. Dis.* 2006, 1:151–70; Taylor, J.P., et al., *Science*, 2002, 296, 1991-1995; Rubinsztein, *Nature*, 2006, 443, 780; Bredeisen, et al., *Nature*, 2006, 443, 796; Spires-Jones, et al., *Acta Neuropathol.*, 2017, 134, 187-205).

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All proteopathies are associated with varying degrees of neuroinflammation. Several mechanisms linking neuroinflammation and proteopathies have been described. For example, extracellular deposition or secretion of protein aggregates can activate immune cells such as microglia and astrocytes, thereby causing a chronic neuroinflammatory state in the brain. In addition, neurons with proteopathy can secrete stress factors or immune molecules which activate immune cells or downregulate immune checkpoints in the brain. Prolonged upregulation of cytokines such as IL-1 β , IL-6 and TNF- α can have harmful effects on the brain and were also found to be able to initiate or aggravate proteinopathy. NDDs are also associated with complement activation, which may lead to increased neuroinflammation and microglial phagocytosis of synapses and neurons. Furthermore, APOE4 – the strongest AD risk factor – is expressed in microglia and astrocytes and increases proteopathy-induced neuroinflammation. Thus, a link between proteinopathy and neuroinflammation has been identified. (Leyns and Holtzman, *Molecular Neurodegeneration*, 2017, 12:50)

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Immunotherapy is an emerging tool in the treatment of NDDs and several antibodies against protein aggregates involved in the pathophysiology of NDDs have been developed and are being investigated in clinical trials. Furthermore, immunotherapy against immune cell surface receptors, secreted immune factors or their cognate receptors have been developed and in several cases investigated in clinical trials against NDDs. However, no cure for any of the NDDs has been found up to now and most clinical trials have failed to show a significant benefit. Hence, there is an urgent need for novel effective therapeutics.

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In therapy of NDDs, the effective delivery of the therapeutic agents into the brain is a major challenge, as passage through the blood brain barrier (BBB) is required. It was found that molecules known as molecular Trojan horses (MTH) bind active transport receptors such as the insulin, transferrin or Low Density Lipoprotein Receptor-related Protein 1 receptor and can enhance entry of therapeutics into the brain by enabling active transport through the BBB.

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Since it has been demonstrated that binding molecules targeted at a protein involved in NDD, such as discussed above, are of clinical value, the development of these therapies has been stimulated. Yet, there is room for improved binding molecules and therapeutic uses thereof.

SUMMARY OF THE INVENTION

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The invention relates to a multispecific binding molecule, preferably an antibody, comprising at least a first binding site binding to a first target selected from the group comprising human tau protein and post-translationally modified human tau protein, such as phosphorylated,

acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, TDP43, mHTT, and fragments thereof and at least a second binding site binding to a second target selected from the group comprising human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, TDP43, mHTT, C1q, C5a, IL1B, IL6, TNF- α , APOE, IL12, IL23 and fragments thereof, wherein the first and second target are different.

Preferably said multispecific binding molecule of the invention, comprises at least a first binding site binding to human tau protein or post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof and at least a second binding site binding to α -synuclein or a fragment thereof.

In another preferred embodiment of the invention the multispecific binding molecule comprises at least a first binding site binding to human tau protein or post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof and at least a second binding site binding to C1q or a fragment thereof.

Further preferred is a multispecific binding molecule of the invention, comprising at least a first binding site binding to α -synuclein or a fragment thereof and at least a second binding site binding to C1q or a fragment thereof.

In another embodiment of the invention, the multispecific binding molecule according to the invention comprises at least one binding site binding to human tau protein selected from the group comprising binding fragments of tau13 and binding fragments of 5A6 and at least one binding site binding to α -synuclein selected from the group comprising binding fragments of Syn211 and binding fragments of H3C.

In yet another embodiment of the invention, the multispecific binding molecule of the invention comprises at least one binding site binding to tau protein selected from the group comprising binding fragments of tau13 and binding fragments of 5A6 and at least one binding site binding to C1q selected from the group comprising binding fragments of JL-1 and binding fragments produced by the hybridomas 23B6C8, 5B5C22, 12A5B7 and 4A4B11.

Further, the invention relates to a multispecific binding molecule comprising at least one binding site binding to C1q selected from the group comprising binding fragments of JL-1 and binding fragments produced by the hybridomas 23B6C8, 5B5C22, 12A5B7 and 4A4B11 and at least one

binding site binding to α -synuclein selected from the group comprising binding fragments of Syn211 and binding fragments of H3C.

In a most preferred embodiment, the multispecific binding molecule of the invention further comprises a moiety that enables shuttling of the binding molecule through the blood brain
5 barrier, preferably wherein said moiety is a binding site.

The multispecific binding molecule of the present invention comprises at least two binding sites binding to at least two proteins or epitopes that are associated with Alzheimer's disease, wherein said at least two proteins or epitopes are different from each other.

The multispecific binding molecule of the present invention comprises at least two
10 binding sites binding to at least two proteins or epitopes that are associated with Lewy Body dementia, wherein said at least two proteins or epitopes are different from each other.

The multispecific binding molecule of the present invention comprises at least two binding sites binding to at least two proteins or epitopes that are associated with Parkinson's Disease, wherein said at least two proteins or epitopes are different from each other.

The multispecific binding molecule of the present invention comprises at least two
15 binding sites binding to at least two proteins or epitopes that are associated with Huntington Disease, wherein said at least two proteins or epitopes are different from each other.

The multispecific binding molecule of the present invention comprises at least two binding sites binding to at least two proteins or epitopes that are associated with Frontotemporal
20 Dementia or Frontotemporal Dementia with Parkinsonism-, wherein said at least two proteins or epitopes are different from each other.

The invention further relates to a multispecific binding molecule that is humanized.

Preferably, the multispecific binding molecule of the invention is bispecific.

More preferably, the multispecific binding molecule according to the invention is
25 trispecific.

In another aspect, the multispecific binding molecule of the invention has a format selected from the group consisting of multispecific binding formats listed in Figure 2 of Brinkmann, et al., MAb, 2017, 9:182-212 and Figure 1 of Spiess, et al.; Molecular Immunology, 2015, 67:95-106, and multispecific antibody conjugates, for example dual-variable-domain (DVD) antibody, trispecific IgG₂ and tetraspecific IgG₂, triple-targeting triplebody, triabody, tribody, trispecific triple heads, trispecific triple dAb, tetraspecific dAb, multispecific dAb, circular dimeric single-chain diabody (CD-scDb), linear dimeric single-chain diabody (LD-scDb), disulfide-stabilized Fv fragment, bis-scFv, tandem tri-scFv, bispecific Fab₂, Fab₃, chemical conjugate trimeric Fab, di-miniantibody, tetrabody, IgG-scFab, scFab-dsscFv, Fv₂-Fc, IgG-scFv fusions, such as Bs1Ab, Bs2Ab, Bs3Ab, Trispecific C-terminal fusion,
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Tri-specific N-terminal fusion, Ts1Ab, Ts2Ab, IgA1, IgA2, IgD, IgE, IgG1, IgG2, IgG3, IgG4, IgM, CODV-Ig, sdAb, trispecific Zybody, tetraspecific Zybody, pentaspecific Zybody, sextaspecific Zybody, septaspecific Zybody, octaspecific Zybody, Knob-into-holes molecules and duobodies

Preferably, the multispecific binding molecule of the invention has the format selected
 5 from the group comprising trispecific triabodies, trispecific tribodies, IgG(H)-scFv, scFv-(H)IgG, IgG(L)-scFv, scFv-(L)IgG, 2scFv-IgG, IgG-2scFv, DVI-IgG, scFab-Fc(kih)-scFv2, scFab-Fc(kih)-scFv, IgG-taFv, scFv4-IgG and TVD-Ig

In a further aspect, the invention relates to a multispecific antibody, wherein the Fc
 10 region of the antibody comprises a mutation at one or more of the following positions: 233, 234, 235, 236, 237, 268, 269, 270, 254, 254, 294, 297, 298, 300, 318, 320, 322, 327, 329, 331.

In another embodiment the invention relates to a multispecific binding molecule
 according to the invention for use in treatment or prevention of neurodegenerative disorders, selected
 from the group comprising Alzheimer's Disease, Lewy Body Dementia, Parkinson's Disease,
 Huntington Disease, Amyotrophic Lateral Sclerosis (ALS), Frontotemporal Dementia, Frontotemporal
 15 Dementia with Parkinsonism-17, Multiple System Atrophy, Corticobasal Degeneration, Progressive
 Supranuclear Palsy, Pick's Disease, Primary Age Related Tauopathy, Argyrophilic Grain Disease and
 Cerebral Amyloid Angiopathy.

Preferably, the invention relates to a multispecific binding molecule for use in treatment
 of Alzheimer's Disease., which preferably can be a bispecific binding molecule for use in the treatment
 20 of Alzheimer's Disease, a trispecific binding molecule for use in the treatment of Alzheimer's disease,
 or a tetraspecific binding molecule for use in the treatment of Alzheimer's disease.

Another aspect of the invention relates to the use of a therapeutic delivery vehicle,
 encoding any of the multispecific binding molecules of the invention in therapy of neurodegenerative
 disorders.

25 Preferably, the invention relates to the use of an adeno-associated virus vector as
 therapeutic delivery vehicle encoding any of the multispecific binding molecules of the invention.

In yet another embodiment, the invention relates to a method for the treatment of neurodegenerative
 disorders, comprising: administration of a multispecific binding molecule according to the invention to
 a subject in need thereof.

30 Preferably, the invention relates to a method for treatment of the neurodegenerative
 disorders, comprising: administration of a nucleic acid construct encoding a multispecific binding
 molecule according to the invention in a therapeutic delivery vehicle to a subject in need thereof.

Further part of the invention is a diagnostic method for the detection of
 neurodegenerative disorders, comprising: adding a multispecific binding molecule according to the
 35 invention to a sample obtained from a subject; determining binding of said binding molecule to any of

the targets selected from human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, , TDP43, mHTT, C1q, C5a, IL1B, IL6, TNF- α , APOE, TREM2, IL12, IL23 and fragments thereof in said sample; diagnosis of neurodegenerative disease if said binding is detected.

The invention also relates to a kit for the diagnosis of neurodegenerative disorders, comprising a multispecific binding molecule according to the invention and means for detection of said binding molecule.

10 LEGENDS TO THE FIGURES

Figure 1 **SDS page to detect bispecific antibodies.** SDS page gel loaded with bispecific antibodies under non-reducing conditions (middle lane) and reducing conditions (right lane). Under non-reducing conditions the bispecific antibodies have the expected molecular weight of approximately 199kDa. Under reducing conditions IgG fragments were observed at expected molecular weights.

15 Figure 2; ELISA results show that bAb0117, a fusion of m9E4 and hu37D3-H9.v28 is able to bind to both recombinant Tau protein and recombinant alpha-synuclein.

Figure 3; ELISA results show that bAb0119, a fusion of AB1 and M1 is able to bind to both recombinant Tau protein and recombinant C1q protein.

20 Figure 4; ELISA results show that bAb0120, a fusion of AB1 and M1 is able to bind to both recombinant Tau protein and recombinant C1q protein

Figure 5; Immunofluorescent staining shows that bAb0117 detects accumulated alpha-synuclein in the cortex of 12 months old Line 61 mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated alpha-synuclein that is phosphorylated at residue 129 - which is a marker for alpha-synuclein pathology and not present in nontransgenic animals.

25 Figure 6; Immunofluorescent staining shows that bAb0117 detects accumulated tau in the hippocampus of 12 months old TMHT mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated tau that is phosphorylated at residue 214 - which is a marker for tau pathology and not present in nontransgenic animals.

30 Figure 7; Immunofluorescent staining shows that bAb0119 detects accumulated tau in the hippocampus of 12 months old TMHT mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated tau that is phosphorylated at residue 214 - which is a marker for tau pathology and not present in nontransgenic animals.

35 Figure 8; Immunofluorescent staining shows that bAb0120 detects accumulated tau in the hippocampus of 12 months old TMHT mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated tau that is phosphorylated at residue 214 - which is a marker for tau pathology.

Figure 9; Results of the Thioflavin T tau aggregation assay.

DETAILED DESCRIPTION

The present invention relates to multispecific binding molecules, preferably antibodies and antigen-binding fragments thereof having specified structural and functional features, and methods of use of the multispecific antibodies and antigen-binding fragments thereof in the diagnosis, prevention or treatment of NDDs.

Target proteins involved in neurodegenerative disorders

The present invention relates to multispecific binding molecules for the prevention, diagnosis or treatment of "neurodegeneration", which refers to the progressive loss of integrity and function of neurons, often leading to cell death. Neurodegeneration, is typically associated with the misfolding of proteins that are abundant in the brain, which leads to "aggregation", e.g. the self-assembly of abnormal proteins to form larger structures and their subsequent "deposition", e.g. precipitation of such aggregates in the brain. Oligomers, as well as insoluble aggregates exhibit neurotoxic activities to the brain tissue through several mechanisms. For example, they can cause oxidative stress, e.g. the generation of reactive oxygen species, resulting in the initiation of a signaling pathway leading to the damaging or apoptosis of cells. In addition, the aggregates contribute to the dysregulation of calcium homeostasis. Both can ultimately lead to synapse and axon dysfunction and death of neurons. Furthermore, these aggregated proteins can be secreted in the extracellular space, leading to neuroinflammation and propagation of protein pathology throughout the brain.

Several proteins were found to be prone to form such aggregates in the brain, and these are therefore suitable targets to prevent or treat neurodegeneration. Treatment of proteopathy-related proteins can be targeted towards multiple forms, including their monomeric, misfolded (conformational) or oligomeric state. Additionally, different aggregated proteins may have overlapping structural features and can be targeted simultaneously with one and the same binding molecule. (Goni, F., et al., J. Neuroinflammation, 2013, 10:914; Zha, J. et al., Scientific reports, 2016, 6:36631) Furthermore, specific post-translational modifications at particular domains can be targeted, such as phosphorylation, acetylation, glycosylation, glycation, prolyl-isomerization, nitration, polyamination, ubiquitination, sumoylation, oxidation and aggregation. Also truncated and cleaved versions of the binding molecule can be targeted. If antibodies are taken up in the cell or expressed in the cell, therapeutics can promote cellular degradation of aggregated proteins. If therapeutics work outside the cell, prevention of their propagation throughout the brain by blocking neuronal uptake or prevention of neuroinflammation could be a mechanisms of action. Additionally, depending on antibody effector function, complexes of therapeutic and aggregated protein can be degraded by immune cells such as microglia or cleared via other pathways. The present invention relates to multispecific binding molecules targeting multiple of the following proteins.

α -Synuclein is abundant in the brain and is predominantly found on presynaptic terminals. It may play a role in the dynamics of presynaptic vesicle release, but its precise function in healthy subjects is not yet clearly understood. However, upon aggregation, toxic soluble oligomers are

formed which can diffuse throughout the neuron and cause cellular damage. Upon their continued aggregation, insoluble fibrils called Lewy Bodies (LB) are formed, which are distinguishable features in Parkinson's disease (PD), Lewy Body Dementia (LBD) and Multiple System Atrophy (MSA). Lewy bodies are also often observed in combination with AD pathology and – if present – contribute to the accelerated disease progression. (Chung et al, JAMA Neurol., 2015, 72, 789–796; Walker et al., Acta Neuropathol., 2015, 129, 729–748; Brenowitz et al., Alzheimers. Dement., 2016, doi:10.1016/j.jalz.2016.09.015; Lemstra et al., J. Neurol. Neurosurg. Psychiatry, 2017, 88, 113–118; Brenowitz et al., Neurology, 2017, doi:10.1212/WNL.0000000000004567; Blanc et al., Alzheimers. Res. Ther., 2017, 9, 47; Irwin et al., Lancet. Neurol., 2017, 16, 55–65; Toledo et al., Acta Neuropathol., 2016, 131, 393–409.

Amyloid- β peptides have about of 36-43 amino acid residues and are derived from the amyloid precursor protein (APP), which can be cleaved by β - and γ -secretases at the N- and C-termini respectively, to form amyloid- β species. These species are prone to oligomerization and subsequent formation and extracellular depositions of toxic plaques. These A β plaques have multiple adverse effects on the function of synapses and axons, including causing oxidative stress, impairing calcium homeostasis and dysregulation of the function of the endoplasmic reticulum (ER) and mitochondria. A β plaques are important for the development of Alzheimer's disease (AD) and cerebral amyloid angiopathy (CAA). Extracellular deposition of amyloid- β is a potent inducer of the activation of microglia and astrocytes. The continued presence of extracellular plaques may therefore create a toxic micro-environment which leads to chronic neuroinflammation in the brain.

Based on these findings, multiple antibodies and small molecules targeting amyloid- β or β -secretase 1 (BACE) have been investigated in clinical trials. However, all failed to show significant clinical benefits.

Tau protein plays an important role in the assembly and stabilization of microtubules in the brain. The interaction of tau protein with tubulin is regulated by dynamic phosphorylation. Under pathological conditions, hyperphosphorylation and aggregation of tau protein occurs. In AD this leads to formation of straight filaments and paired helical filaments, which subsequently form the neurofibrillary tangles and neuropil threads found in patient brains. In other tauopathies, these aggregates can have different structural features and affect also other cell types, such as astrocytes. As with α -synuclein, tau aggregation therefore leads to both loss-of-function and gain-of-toxic-function, both contributing to neurodegeneration.

Furthermore, several independent studies show that tau immunization not only inhibits tau pathology in mice, but simultaneously enhanced clearance of amyloid- β plaques. It was hypothesized that the reduction in A β plaques is due to a reduction in APP synthesis and/or amyloidogenic processing as a result of immunization. Moreover, antibody treatment could induce activation of microglia which facilitates A β clearance. (Rajamohamedsait, H. et al, Scientific reports, 2017, 7:17034; Castillo-Carranza, C.M. et al, J. Neurosci, 2015, 35(12):4857– 4868; Dai, C., et al., Alzheimers Research and Therapy, 2017, 9:1)

Examples of “tauopathies”, e.g. class of NDDs associated with the hyperphosphorylation and subsequent aggregation of tau proteins in the brain, are corticobasal degeneration (CBD), frontotemporal dementia (FTD) or Pick’s disease (PiD), frontotemporal dementia with parkinsonism linked to chromosome 17 (FTDP-17), progressive supranuclear palsy (PSP), and chronic traumatic encephalopathy (CTE). Alzheimer’s disease (AD) is classified as a secondary tauopathy, since amyloid- β is also additionally present. Tau pathology is strongly linked to cellular dysfunction, synapse loss and neurodegeneration and the symptoms of tauopathies therefore correspond to the affected anatomical regions. Additionally, tau pathology is often found in synucleinopathies, HD, ALS and other NDDs.

TAR DNA-binding protein 43 (TDP-43) is a nuclear protein that binds both RNA and DNA and is involved in regulating splicing, trafficking and stabilization of RNA, as well as miRNA production. In neurodegenerative disorders TDP-43 becomes mislocalized in the cytoplasm where it aggregates, forming stress granules and insoluble inclusion bodies. TDP-43 inclusions are observed in patients with Frontotemporal Dementia (FTD) or *Amyotrophic Lateral Sclerosis (ALS)*. In addition, TDP-43 pathology is observed in a quarter of the patients with AD. (Montine et al., *Acta Neuropathol.*, 2012, 123, 1–11; Takeda, T., *Neuropathology*, 2018; 38, 72–81, Josephs et al., *Neurology*, 2008, 70, 1850–1857; Josephs et al., *Acta Neuropathol.*, 2014, 127, 441–450; Behrouzi et al., *Acta Neuropathol. Commun.*, 2016, 4, 33).

The mutant Huntingtin protein (mHTT) is a mutant of the huntingtin gene caused by an expansion of the polyglutamine repeat within exon 1 of the huntingtin gene on chromosome 4, exceeding 35 CAG repeats. The mutant has been linked to the onset and progression of Huntington’s disease (HD), an inherited disorder characterized by the neuronal dysfunction and degeneration in the striatum and cerebral cortex. Although the exact mechanism behind HD has not been fully elucidated, it is clear that mHTT alters intracellular Ca^{2+} homeostasis, disrupts intracellular trafficking and impairs gene transcription, processes that induce neurodegeneration. In contrast to for example sporadic AD and PD, HD is a genetic disease and the pathology is expected to be solely the results of mHTT dysfunction. Interestingly however, HD frequently features aggregates composed of pathological tau, α -synuclein and TDP-43 (St-Amour, I, et al., *Acta Neuropathol.* 2017, <https://doi.org/10.1007/s00401-017-1786-7>, Fernandez-Nogales et al., *Nat Med*, 2014, 20, 881–885; Vuono et al., *Brain*, 2015, 138, 1907–1918).

“Neuroinflammation”, e.g. the inflammation of nervous tissue, has been linked to neurodegeneration. Pathological protein aggregates directly and indirectly activate microglia and astrocytes and may initially lead to successful phagocytosis of these proteins. However, under chronic conditions these cells secrete proinflammatory cytokines, causing harmful neuroinflammation and neurodegeneration. These cytokines are additionally involved in the activation of neuronal intracellular pathways (e.g. kinases that phosphorylate tau proteins), which induces misfolding and aggregation of NDD related proteins. These pro-inflammatory cytokines may also lead to further neuroinflammation and further aggravate neurodegeneration. Microglia can for example induce a neurotoxic phenotype in astrocytes by a combination of IL1 α , TNF- α and C1q (Liddelow, S.A., et al.,

Nature, 2017, 541, 481–487). Moreover, activated microglia increase tau spreading, thereby contributing to the progression of intracellular protein aggregates throughout the brain. (Maphis, N., et al., *Brain*, 2015, 138 (6):1738-55; Asai, H., et al., *Nature Neuroscience*, 2015, 18, 1584–1593). Secreted immune factors are attractive targets for therapy as they are available in the extracellular space. Their action can be neutralized by therapeutics binding directly to these immune factors or by blocking their cognate receptors. Accordingly, complement proteins and cytokines, such as C1q, C5a, IL1 β , IL6, TNF- α , and IL12/IL23 play an important role in the pathobiology of NDDs.

For example, Interleukin (IL)-1 β was found to be consistently upregulated in AD, related tauopathies, PD, HD and various proteopathy animal models of these disorders. Cacabelos et al., *Methods Find. Exp. Clin. Pharmacol.*, 1991, 13, 455–458; Cacabelos et al., *Methods Find. Exp. Clin. Pharmacol.* 1994, 16, 141–151; Gitter et al., *Proc. Natl. Acad. Sci. U. S. A.*, 1995, 92, 10738–10741; Blum-Degen et al., *Neurosci. Lett.*, 1995, 202, 17–20; Mogi et al., *Neurosci. Lett.*, 1994, 180, 147–150; Mogi et al., *Neurosci. Lett.*, 1996, 211, 13–16; King et al., *Alzheimer Dis. Assoc. Disord.*, 2017, doi:10.1097/WAD.0000000000000211; Björkqvist et al., *J. Exp. Med.*, 2008, 205, 1869–1877; Ona et al., *Nature*, 1999, 399, 263–267; Cook et al., *Hum. Mol. Genet.*, 2015, 24, 6198–6212). This proinflammatory cytokine is expressed by several cell types in the brain, though primarily by microglia as a result of insult or injury. Interestingly, IL-1 β was found to enhance A β phagocytosis by microglia, thereby reducing A β plaques in the brain, which suggests a protective role of IL-1 β against neurodegeneration in early stages of AD. However, several studies also suggest induction of tau pathology by IL-1 β . Although the exact underlying mechanism has not yet been elucidated, there is convincing evidence that IL-1 β is in fact able to mediate neuronal kinase activity, thereby stimulating phosphorylation and subsequent aggregation of tau. Indeed, several studies find opposite effects of immune pathways on A β on the one hand, and synapse loss and tau pathology on the other hand. (Leyns and Holtzman, *Molecular Neurodegeneration*, 2017, 12:50) Administration of an IL1R (the shared receptor for IL1A and IL1B) monoclonal antibody in a mouse model with combined A β and tau pathology has favorable effects on inflammation, cognition, A β and tau pathology (Kitazawa, M, et al., *J Immunol*, 2011; 187:6539-6549)

IL-6 is also upregulated in AD, related tauopathies, PD, HD and various proteopathy animal models of these disorders. (Luterman et al. *Arch. Neurol.* 2000, 57, 1153–1160; Jiang et al., *Neurobiol. Aging*, 2015, 36, 3176–3186; Jiang et al., *Neuropharmacology*, 2016, 105, 196–206; Kovac et al., *J. Immunol.*, 2011, 187, 2732–2739; Khandelwal et al., *Mol. Cell. Neurosci.*, 2012, 49, 44–53; Cook et al., *Hum. Mol. Genet.*, 2015, 24, 6198–6212; Mravec et al., *J. Neuroinflammation*, 2016, 13, 15; Blum-Degen et al., *Neurosci. Lett.*, 1995, 202, 17–20; Mogi et al., *Neurosci. Lett.*, 1994, 180, 147–150; Mogi et al., *Neurosci. Lett.*, 1996, 211, 13–16; King et al., 2017, doi:10.1097/WAD.0000000000000211; Björkqvist et al., *J. Exp. Med.*, 2008, 205, 1869–1877; Hull et al., *Neurobiol. Aging*, 1996, 17, 795–800; Cojocaru et al., *Rom. J. Intern. Med.* 2011, 49, 55–58; Rojanathammanee et al., *J. Neuroinflammation*, 2011, 8, 44; Silvestroni et al, *Neuroreport*, 2009, 20, 1098–1103. Similarly to IL-1 β , overexpression of IL-6 was correlated with a reduction of A β plaques in the brain, whilst tau phosphorylation was increased through activation of the kinases p38 and cdk5.

A correlation between increased serum levels of IL-6 and parkinsonism has also been established. (Leyns and Holtzman, *Molecular Neurodegeneration*, 2017, 12:50)

The cytokines IL-12 and IL-23 were found to play a significant role in the pathology of AD and the inhibition of IL-12/IL-23 (which share the P40 subunit) signaling pathway as well as the administration of neutralizing antibodies resulted in a reduction of A β plaques in the brain. (vom Berg, et. al., *Nat. Med.*, 2012, 18, 1812-1819)

Tumor necrosis factor alpha (TNF- α) is another proinflammatory cytokine that was correlated to tau and A β pathologies and consequent degeneration of neurons. (Jiang et al., *Neurobiol. Aging*, 2015, 36, 3176–3186; Jiang et al., *Neuropharmacology*, 2016, 105, 196–206; Kovac et al., *J. Immunol.*, 2011, 187, 2732–2739; Khandelwal et al., *Mol. Cell. Neurosci.*, 2012, 49, 44–53; Cook et al., *Hum. Mol. Genet.*, 2015, 24, 6198–6212; Mravec et al., *J. Neuroinflammation*, 2016, 13, 15; Novak et al., *Cell. Mol. Neurobiol.*, 2017, doi:10.1007/s10571-017-0491-3; Combs et al., *J. Neurosci.*, 2001, 21, 1179–1188; Perry et al., *Neurobiol. Aging*, 2001, 22, 873–883; King et al., 2017, doi:10.1097/WAD.0000000000000211. Szejder-Pacholek et al., *Pharmacol. Rep.* 69, 2017, 242–251; Hsiao et al., *Hum. Mol. Genet.*, 2014, 23, 4328–4344; Björkqvist et al. *J. Exp. Med.* 2008, 205, 1869–1877). For example, it was found that A β can bind a receptor of TNF- α (TNFR1), eventually leading to activation of Nuclear Factor κ B (NF- κ B) and neuronal apoptosis. In addition, upregulation of TNF- α was found to stimulate the expression of other local inflammatory mediators, ultimately leading to increased levels of A β and hyperphosphorylated tau. Finally, it was shown that TNF- α can eventually lead to apoptosis in neurons, amongst others through activation of caspases. (Leyns and Holtzman, *Molecular Neurodegeneration*, 2017, 12:50) Likewise, TNF γ and TNF type 1 were linked to the pathology of AD and PD. Several studies find favorable effects of targeting TNF- α and its receptors in mouse models of AD and some compounds are currently in clinical trials (Chang, R. et al., *J Cent Nerv Syst Dis*, 2017, 9: 1–5)

Complement is a potent pro-inflammatory system and one of the major pathways of the innate immune system. The complement system is crucial in the first line of defense against microbes and pathogens, but may also cause damage to self through recruitment of immune cells and formation of the membrane attack complex (MAC). It is comprised of many proteins that lead to a cascade of events, ultimately forming highly inflammatory peptides such as complement component (C) 5a. Therapies targeting C5a were shown to be beneficial in animals models of AD, PD and HD (Landlinger et al., *J. Neuroinflammation*, 2015, 12, 150; Gordon et al., *FASEB J.*, 2015, 29; Woodruff et al., *FASEB J.*, 2006, 20, 1407–1417). Interestingly, a correlation between upregulation of C1q (the first subcomponent of the C1 complex) and multiple proteinopathies has been established. C1q is often upregulated in the striatum of HD patients, substantia nigra of PD and decorates both amyloid plaques and neurofibrillary tangles in AD brains. (McGeer et al., *Neurosci. Lett.*, 1989, 107, 341–346; Rogers et al., *Proc. Natl. Acad. Sci.*, 1992, 89, 10016–10020; Afagh et al., *Exp. Neurol.*, 1996, 138, 22–32; Shen et al., *Neurosci. Lett.*, 2001, 305, 165–168; Schwab et al., *Brain Res.*, 1996, 707, 196–205; Depboylu et al., *J. Neuropathol. Exp. Neurol.*, 2011, 70, 125–132; Singhrao et al., *Exp. Neurol.*, 1999, 159, 362–376). Multiple mouse models of Alzheimer's disease indicated upregulation of C1q prior to

plaque formation. In addition, C1q-tagged synapses can be phagocytosed by microglia in a pathway involving C3 and complement receptor 3 (CR3). This pathway plays an important role in neurodevelopment but can be induced in the adult brain under a wide range of pathological conditions such as virus infection and neurodegeneration. Early synapse loss was prevented by infusion of an anti-C1q monoclonal antibody in mouse models of AD, and an immunotherapeutic is currently in clinical trials. (Hong, S. et al., *Science*, 2016, 352, 712–716). C1q deposition leads to early loss of synapses via the C3-CR3 pathway. Furthermore, with increased neurodegeneration or BBB breakdown, C1q deposition also leads to induction of other downstream components of the classical complement pathway. This causes recruitment of immune cells and neuroinflammation, formation of the MAC and ultimately leads to neurodegeneration. Targeting C1q with immunotherapy may therefore diminish early synapse loss, neuroinflammation, MAC formation and ultimately neurodegeneration in AD and other tauopathies, synucleinopathies and HD. Morgan, B.P., *Semin Immunopathol.*, 2017, 1-12. Morgan, B.P., *Semin. Immunopathol.*, 2018, 40:113-124.

The gene encoding Apolipoprotein E (APOE), most notably APOE4, has been identified to be the strongest risk factor for late-onset AD and was additionally correlated to other proteopathies, such as HD, DLB and PD. (Strittmatter et al., *Proc. Natl. Acad. Sci. U. S. A.*, 1993, 90, 1977–1981; Holtzman et al., *Proc. Natl. Acad. Sci. U. S. A.*, 2000, 97, 2892–2897; Lambert et al., *Nat. Genet.*, 2009, 41, 1094–1099; Kalman et al., *Neurobiol. Aging*, 2000, 21, 555–558; Wakabayashi et al., *Acta Neuropathol.*, 1998, 95, 450–454; Tsuang et al., *Neurology* 2005, 64, 509–513, Pankratz et al., *Mov. Disord.* 2006, 21, 45–49; Tsuang et al., *JAMA Neurol.*, 2013, 70, 223–228, Panas et al., *J. Neurol.* 1999, 246, 574–577; Kehoe et al., *J. Med. Genet.*, 1999, 36, 108–111; Rita Guerreiro et al., *Lancet Neurol.*, 2018, 17, 64-74). For example, it was demonstrated that APOE influences deposition of A β plaques in the brain. In addition, a mouse model using P301S tau transgenic mice, indicated both increased levels of phosphorylated tau in the brain, as well as increased cytokine levels, such as IL-1 β , IL-1 α and TNF- α for P301S/E4 mice, compared to P301S/E2 and P301S/E3 mice, whereas these changes were largely absent in P301S/KO mice. Thus, APOE plays a prominent role in the aggravation of neurodegeneration both through mediation of aggregation, as well as by inducing neuroinflammation through increased innate immunity. (Yang Shi et al., *Nature*, 2017, 549, 523). Anti-APOE immunotherapy was shown to strongly reduce plaque load and behavioral deficits in mice: (Kim, J., *J. Exp. Med.*, 2012, 209 (12) 2149-2156; Liao, F., *J Neuroscience*, 2014, 34 (21) 7281-7292). Immunotherapy specifically directed towards the APOE4 allele decreased amyloid load, hyperphosphorylated tau and behavioral deficits in transgenic animals (Luz, I., *Current Alzheimer's Research*, 2016, 13 (8), 918-929).

“Neurodegenerative disorders” (NDDs), e.g. disorders that are characterized by neurodegeneration, are typically characterized by the accumulation of aggregates, formed by one or multiple proteins mentioned above, in the brain. In most cases, the underlying cause of the NDD is unknown, although certain risk factors have been identified, such as old age, (multiple) head trauma or genetic predisposition.

Arguably the most common and well-known NDD is Alzheimer's Disease (AD). Symptoms include impaired memory, disorientation and behavioral changes. It is associated with aggregates composed of hyperphosphorylated tau protein, forming neurofibrillary tangles in the neuron, and accumulation of extracellular plaques formed from amyloid β . In addition, aggregation of TDP-43 and α -synuclein have also been observed in AD patients.

Another NDD associated with accumulation of A β is cerebral amyloid angiopathy (CAA), in which A β aggregates cause multiple strokes in the brain, which can lead to paralysis, dementia or even death.

Chronic traumatic encephalopathy (CTE) is a tauopathy that is caused by multiple traumatic injuries to the brain. Traumatic brain injury can result in amyloid β plaques and tau, TDP-43 and α -synuclein inclusions. (Kenney, K., et al., J Neuropathol Exp Neurol. 2018 Jan 1;77(1):50-63.) Most patients are former athletes (of contact sports such as boxing or ice-hockey) or military veterans.

Parkinson's Disease (PD) is a NDD that affects the dopaminergic neurons in the substantia nigra, which mainly affects the motor system. Symptoms include tremor, slowness of moving, rigidity and difficulty of walking. Although the underlying cause of PD remains largely unclear, Lewy Bodies (e.g. aggregates of α -synuclein) in the nerve cells have been observed.

Multiple System Atrophy (MSA) or Shy-Drager syndrome, is a rare NDD that shows clinical overlap with PD, but shows little response to dopamine agonists that are often used in the treatment of PD. Like PD, MSA has been linked to the formation of the Lewy Bodies in the brain.

Lewy Body Dementia (LBD) is a form of dementia that is characterized by the formation of the Lewy Bodies in the brain. It shares symptoms with PD and AD.

Corticobasal degeneration (CBD) and progressive supranuclear palsy (PSP) are two other progressive NDDs that are associated with accumulation of abnormal tau aggregates. Both disorders are primarily characterized with moving dysfunction and symptoms are observed similar to PD, but cognitive and/or behavioral abnormalities have also been observed.

Huntington Disease (HD) is a genetic disorder caused by a mutation in the gene coding for the Huntington protein. Expansion of the CAG repeats in the gene results in an abnormal protein that damages brain cells, affecting movement, behavior and cognition. In most cases, the on-set of the disease is between 30-40 years of age of the patient.

Amyotrophic Lateral Sclerosis (ALS) is a motor-neuron disease, characterized by the gradually worsening of muscle weakness, which leads to difficulties in moving, speaking, swallowing and respiratory failure. In most cases, deposition of TDP-43 in the cytoplasm is observed which impairs RNA processing.

Frontotemporal Dementia (FTD) or Pick's disease is another form of dementia which is characterized by aggregations of tau protein and/or TDP-43 in the frontal and temporal lobes of the brain. These areas of the brain are associated with language (temporal) and behavior (frontal) and

consequently symptoms are indeed primarily correlated with changes in personality, behavior and speech. This type in dementia is often observed in relatively young patients (40-60) years of age.

In approximately 20% of the cases a genetic mutation is the underlying cause of the disease. For example, Tau-positive frontotemporal dementia with parkinsonism (FTDP-17) is caused
5 by a mutation in the MAPT gene that encodes for tau protein and is a variant of FTD, in which the patient exhibits features related to movement disorders.

Multispecific binding molecules

Many NDDs are associated with mixed pathologies of different disease targets. For example AD is characterized by both A β plaques, as well as aggregates of phosphorylated tau. In
10 addition, aggregates of α -synuclein and TDP-43 or co-aggregates thereof (for example with tau or Amyloid β) are also frequently observed in patients suffering from AD. Aggregates comprised of different proteins have also been observed. Such mixed pathology is partly the result of the ability of aggregates to interact with one another and induce aggregation of other proteins. However, in the aging brain multiple types of pathology are often observed. Aging and NDDs may therefore make the
15 brain more susceptible to the accumulation of various aggregates. In some disorders, mixed pathologies are in fact a prerequisite for full manifestation of the disease. For example, tau pathology does not progress to the neocortex in absence of A β plaques. Additionally, the presence of Lewy bodies has been associated with worse disease outcomes in AD patients. Interestingly, with advancing age the amount of mixed pathology increases, including in pre-symptomatic people. As the world
20 population is increasingly aging, the amount of people with mixed pathology is expected to increase substantially in the future.

In addition, synergies between proteins involved in proteopathies and neuroinflammation have also been demonstrated. For example, A β plaques in AD induce microglia activation, which subsequently initiate aggregation of tau proteins, hence enhancing disease progression.

25 In addition, synergies can exist in immune pathways. For example, TNF- α can induce microglial activation and associated neuroinflammation. This may lead to secretion of more TNF- α and other cytokines.

Immune pathways can also modulate the relationship between proteopathy and cytokine secretion. For example, APOE genotype alters pathological A β or pathological tau-induced microglia
30 and astrocyte activation and subsequent secretion of pro-inflammatory cytokines (e.g. IL1 β and TNF- α) and their downstream effects.

Immune molecules can also mediate some of the toxic effects of proteopathy. Aggregated proteins or cells containing aggregated proteins in tauopathies, synucleinopathies and HD have been shown to induce classical complement activation. A β can for example induce microglial phagocytosis
35 of synapses in a pathway involving complement-involved proteins C1q, C3 and CR3. (Hong, S. et al., Science, 2016, 352, 712–716).

Immune cells such as microglia can also use combinations of immune factors (IL1 α , TNF- α and C1q) to induce reactive neurotoxic astrocytes (Liddelow, S.A., et al., Nature, 2017, 541, 481–487).

5 The current invention thus relates to the use of multispecific binding molecules, specifically binding at least two different molecules of the abovementioned target for therapy or diagnosis of NDDs.

Herein, the use of multispecific binding molecules offers an additional advantage over the use of multiple monospecific binding molecules. For example, the efficacy of therapy of NDDs is drastically increased when multispecific binding molecules are used instead of a combination of
10 multiple monospecific binding molecules. It allows the targeting of several pathological proteins simultaneously, whereas in a monospecific binding molecule approach multiple binding molecules are required to achieve a similar therapeutic effect. Hence, the total concentration of the multispecific binding molecule may remain relatively low, which is associated with a decreased risk of adverse effects, fewer clinical difficulties and lower production costs. Additionally, unknown interactions,
15 differences in brain uptake or clearance kinetics between different molecules may be a limitation in combining multiple monospecific binding molecules.

Accordingly, the present invention relates to multispecific binding molecules and the use thereof. The present invention is directed at multispecific binding molecules, comprising at least a first binding site binding to a first target selected from the group comprising human tau protein and
20 post-translationally modified, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated, aggregated, cleaved and truncated versions thereof, α -synuclein, β -amyloid, TDP43, mHTT and fragments thereof, and a second binding site binding to a second target selected from the group comprising human tau protein and post-translational modified, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-
25 isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated, aggregated, cleaved and truncated versions thereof, α -synuclein, β -amyloid, TDP43, mHTT, C1q, C5a, IL1 β , IL6, TNF- α , APOE, IL12/IL23 and fragments thereof, wherein the first and second target is different. In some instances, the present invention relates to multispecific binding molecules having a further binding site that binds to a third target selected from the group comprising human tau protein and post-
30 translational modified, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated, aggregated, cleaved and truncated versions thereof, α -synuclein, β -amyloid, TDP43, mHTT, C1q, C5a, IL1 β , IL6, TNF- α , APOE, IL12/IL23 and fragments thereof, wherein these targets are different. Many combinations are possible and accordingly the present invention comprises a plurality of different multispecific binding
35 molecules, including but not limited to the combinations listed below.

Table 1; First group of targets

Entry	Target
A1	tau protein targets
A2	α -synuclein
A3	β -amyloid
A4	TDP43
A5	mHTT

Table 2; Second group of targets

Entry	Target
B1	C1q
B2	C5a
B3	IL1 β
B4	IL6
B5	TNF- α
B6	APOE
B7	IL12
B8	IL23

10 Multispecific binding molecules having at least one of the following binding specificities are included in the present invention, where A1-A5 and B1-B8 are indicated in the Tables 1-2 above

15 A1+A2, A1+A3, A1+A4, A1+A5, A2+A3, A2+A4, A3+A4, A4+A5, A1+B1, A1+B2, A1+B3, A1+B4,
A1+B5, A1+B6, A1+B7, A1+B8, A2+B1, A2+B2, A2+B3, A2+B4, A2+B5, A2+B6, A2+B7, A2+B8,
A3+B1, A3+B2, A3+B3, A3+B4, A3+B5, A3+B6, A3+B7, A3+B8, A4+B1, A4+B2, A4+B3, A4+B4,
A4+B5, A4+B6, A4+B7, A4+B8, A5+B1, A5+B2, A5+B3, A5+B4, A5+B5, A5+B6, A5+B7, A5+B8,
A1+A2+A3, A1+A2+A4, A1+A3+A4, A1+A4+A5, A2+A3+A4

20	A1+A2+B1,	A1+A2+B2,	A1+A2+B3,	A1+A2+B4,	A1+A2+B5,	A1+A2+B6,	A1+A2+B7,	A1+A2+B8,
	A1+A3+B1,	A1+A3+B2,	A1+A3+B3,	A1+A3+B4,	A1+A3+B5,	A1+A3+B6,	A1+A3+B7,	A1+A3+B8,
	A1+A4+B1,	A1+A4+B2,	A1+A4+B3,	A1+A4+B4,	A1+A4+B5,	A1+A4+B6,	A1+A4+B7,	A1+A4+B8,
	A1+A5+B1,	A1+A5+B2,	A1+A5+B3,	A1+A5+B4,	A1+A5+B5,	A1+A5+B6,	A1+A5+B7,	A1+A5+B8,
	A2+A3+B1,	A2+A3+B2,	A2+A3+B3,	A2+A3+B4,	A2+A3+B5,	A2+A3+B6,	A2+A3+B7,	A2+A3+B8,
	A2+A4+B1,	A2+A4+B2,	A2+A4+B3,	A2+A4+B4,	A2+A4+B5,	A2+A4+B6,	A2+A4+B7,	A2+A4+B8,
25	A3+A4+B1,	A3+A4+B2,	A3+A4+B3,	A3+A4+B4,	A3+A4+B5,	A3+A4+B6,	A3+A4+B7,	A3+A4+B8,
	A4+A5+B1,	A4+A5+B2,	A4+A5+B3,	A4+A5+B4,	A4+A5+B5,	A4+A5+B6,	A4+A5+B7,	A4+A5+B8,
	A1+B1+B2,	A1+B1+B3,	A1+B1+B4,	A1+B1+B5,	A1+B1+B6,	A1+B1+B7,	A1+B1+B8,	A1+B2+B3,
	A1+B2+B4,	A1+B2+B5,	A1+B2+B6,	A1+B2+B7,	A1+B2+B8,	A1+B3+B4,	A1+B3+B5,	A1+B3+B6,
	A1+B3+B7,	A1+B3+B8,	A1+B4+B5,	A1+B4+B6,	A1+B4+B7,	A1+B4+B8,	A1+B5+B6,	A1+B5+B7,
	A1+B5+B8,	A1+B6+B7,	A1+B6+B8,	A1+B7+B8,	A2+B1+B2,	A2+B1+B3,	A2+B1+B4,	A2+B1+B5,
30	A2+B1+B6,	A2+B1+B7,	A2+B1+B8,	A2+B2+B3,	A2+B2+B4,	A2+B2+B5,	A2+B2+B6,	A2+B2+B7,
	A2+B2+B8,	A2+B3+B4,	A2+B3+B5,	A2+B3+B6,	A2+B3+B7,	A2+B3+B8,	A2+B4+B5,	A2+B4+B6,
	A2+B4+B7,	A2+B4+B8,	A2+B5+B6,	A2+B5+B7,	A2+B5+B8,	A2+B6+B7,	A2+B6+B8,	A2+B7+B8,
	A3+B1+B2,	A3+B1+B3,	A3+B1+B4,	A3+B1+B5,	A3+B1+B6,	A3+B1+B7,	A3+B1+B8,	A3+B2+B3,
	A3+B2+B4,	A3+B2+B5,	A3+B2+B6,	A3+B2+B7,	A3+B2+B8,	A3+B3+B4,	A3+B3+B5,	A3+B3+B6,
	A3+B3+B7,	A3+B3+B8,	A3+B4+B5,	A3+B4+B6,	A3+B4+B7,	A3+B4+B8,	A3+B5+B6,	A3+B5+B7,
35	A3+B5+B8,	A3+B6+B7,	A3+B6+B8,	A3+B7+B8,	A4+B1+B2,	A4+B1+B3,	A4+B1+B4,	A4+B1+B5,
	A4+B1+B6,	A4+B1+B7,	A4+B1+B8,	A4+B2+B3,	A4+B2+B4,	A4+B2+B5,	A4+B2+B6,	A4+B2+B7,

A4+B2+B8, A4+B3+B4, A4+B3+B5, A4+B3+B6, A4+B3+B7, A4+B3+B8, A4+B4+B5, A4+B4+B6, A4+B4+B7, A4+B4+B8, A4+B5+B6, A4+B5+B7, A4+B5+B8, A4+B6+B7, A4+B6+B8, A4+B7+B8, A5+B1+B2, A5+B1+B3, A5+B1+B4, A5+B1+B5, A5+B1+B6, A5+B1+B7, A5+B1+B8, A5+B2+B3, A5+B2+B4, A5+B2+B5, A5+B2+B6, A5+B2+B7, A5+B2+B8, A5+B3+B4, A5+B3+B5, A5+B3+B6, A5+B3+B7, A5+B3+B8, A5+B4+B5, A5+B4+B6, A5+B4+B7, A5+B4+B8, A5+B5+B6, A5+B5+B7, A5+B5+B8, A5+B6+B7, A5+B6+B8, A5+B7+B8.

In a preferred embodiment the multispecific binding molecule have the following binding specificities: A1+A2, A1+B1, A2+B1.

Additionally, binding molecules have been identified that recognize a common amyloid fold that is shared by multiple toxic misfolded proteins. For example, the general amyloid interaction motif (GAIM) is a fragment of a capsid protein and was found to simultaneously bind α -synuclein, tau protein and amyloid- β . Such binding molecules can also form a binding site of a multispecific binding molecule.

In a preferred embodiment the multispecific binding molecule is a multispecific antibody.

The present invention includes antibodies and methods of use thereof. As used herein, the term "antibody" refers to any form of antibody that exhibits the desired biological activity. Thus, it is used in the broadest sense and specifically covers, but is not limited to, monoclonal antibodies (including full length monoclonal antibodies comprising two light chains and two heavy chains), polyclonal antibodies, humanized antibodies, fully human antibodies, chimeric antibodies and camelized single domain antibodies.

The present invention includes antigen-binding fragments and methods of use thereof. As used herein, unless otherwise indicated, "antibody fragment" or "antigen-binding fragment" refers to antigen-binding fragments of antibodies, *i.e.* antibody fragments that retain the ability to bind specifically to the antigen bound by the full-length antibody, *e.g.* fragments that retain one or more CDR regions. Examples of antigen-binding fragments include, but are not limited to, Fab, Fab', F(ab')₂, and Fv fragments; diabodies; linear antibodies; single-chain antibody molecules, *e.g.*, sc-Fv; nanobodies and multispecific antibodies formed from antibody fragments.

The present invention includes Fab fragments and methods of use thereof. A "Fab fragment" is comprised of one light chain and the C_{H1} and variable regions of one heavy chain. The heavy chain of a Fab molecule cannot form a disulfide bond with another heavy chain molecule. A "Fab fragment" can be the product of papain cleavage of an antibody.

The present invention includes antibodies and antigen-binding fragments thereof which comprise an Fc region and methods of use thereof. An "Fc" region contains two heavy chain fragments comprising the C_{H2} and C_{H3} domains of an antibody. The two heavy chain fragments are held together by two or more disulfide bonds and by hydrophobic interactions of the C_{H3} domains.

The present invention includes Fab' fragments and methods of use thereof. A "Fab' fragment" contains one light chain and a portion or fragment of one heavy chain that contains the V_H

domain and the C_{H1} domain and also the region between the C_{H1} and C_{H2} domains, such that an interchain disulfide bond can be formed between the two heavy chains of two Fab' fragments to form a F(ab')₂ molecule.

5 The present invention includes F(ab')₂ fragments and methods of use thereof. A "F(ab')₂ fragment" contains two light chains and two heavy chains containing a portion of the constant region between the C_{H1} and C_{H2} domains, such that an interchain disulfide bond is formed between the two heavy chains. A F(ab')₂ fragment thus is composed of two Fab' fragments that are held together by a disulfide bond between the two heavy chains. A "F(ab')₂ fragment" can be the product of pepsin cleavage of an antibody.

10 The present invention includes Fv fragments and methods of use thereof. The "Fv region" comprises the variable regions from both the heavy and light chains, but lacks the constant regions.

The present invention includes scFv fragments and methods of use thereof. The term "single-chain Fv" or "scFv" antibody refers to antibody fragments comprising the V_H and V_L domains of an antibody, wherein these domains are present in a single polypeptide chain. Generally, the Fv polypeptide further comprises a polypeptide linker between the V_H and V_L domains which enables the scFv to form the desired structure for antigen-binding. For a review of scFv, see Pluckthun (1994) The Pharmacology of Monoclonal Antibodies, vol. 113, Rosenberg and Moore eds. Springer-Verlag, New York, pp. 269-315. See also, International Patent Application Publication No. WO 88/01649 and U.S. Pat. Nos. 4,946, 778 and 5,260,203.

20 The present invention includes domain antibodies and methods of use thereof. A "domain antibody" is an immunologically functional immunoglobulin fragment containing only the variable region of a heavy chain or the variable region of a light chain. In some instances, two or more V_H regions are covalently joined with a peptide linker to create a bivalent domain antibody.

25 The present invention includes multivalent antibodies and methods of use thereof. A "multivalent antibody" comprises at least two antigen-binding sites. These binding sites can have the same antigen specificity, or can bind different specificities. In the latter case, the multivalent antibody is at least bispecific. A multivalent antibody can also be at least bivalent for one target and at least bivalent for a further target. In that case the antibody is multispecific and multivalent.

30 The present invention includes camelized single domain antibodies and methods of use thereof. In certain embodiments, antibodies herein also include camelized single domain antibodies. See, e.g., Muyldermans et al. Trends Biochem. Sci. 2001, 26:230; Reichmann et al. J. Immunol. Methods 1999, 231:25; WO 94/04678; WO 94/25591; U.S. Pat. No. 6,005,079).

In one embodiment, the present invention provides single domain antibodies comprising two V_H domains with modifications such that single domain antibodies are formed.

35 The present invention includes diabodies and methods of use thereof. As used herein, the term "diabodies" refers to small antibody fragments with two antigen-binding sites, which fragments comprise a heavy chain variable domain (V_H) connected to a light chain variable domain (V_L) in the

same polypeptide chain (V_H - V_L or V_L - V_H). By using a linker that is too short to allow pairing between the two domains on the same chain, the domains are forced to pair with the complementary domains of another chain and create two antigen-binding sites. Diabodies are described more fully in, e.g., EP 404,097; WO 93/11161; and Holliger et al. Proc. Natl. Acad. Sci. USA 1993, 90: 6444-6448.

5 Various other forms of multispecific antibodies are known. All multispecific binding formats listed in Figure 2 of Brinkmann, et al., MAbs, 2017, 9:182-212 and Figure 1 of Spiess, et al.; Molecular Immunology, 2015, 67:95-106 are possible formats used for multispecific binding molecules of the present invention. Included herein are also multispecific antibody conjugates, for example dual-
 10 variable-domain (DVD) antibody, trispecific IgG₂ and tetraspecific IgG₂, triple-targeting triplebody, triabody, tribody, trispecific triple heads, trispecific triple dAb, tetraspecific dAb, multispecific dAb, circular dimeric single-chain diabody (CD-scDb), linear dimeric single-chain diabody (LD-scDb), disulfide-stabilized Fv fragment, bis-scFv, tandem tri-scFv, bispecific Fab2, Fab3, chemical conjugate trimeric Fab, di-miniantibody, tetrabody, IgG-scFab, scFab-dsscFv, Fv2-Fc, IgG-scFv fusions, such as Bs1Ab, Bs2Ab, Bs3Ab, Trispecific C-terminal fusion, Tri-specific N-terminal fusion, Ts1Ab, Ts2Ab,
 15 IgA1, IgA2, IgD, IgE, IgG1, IgG2, IgG3, IgG4, IgM, CODV-Ig, sdAb, trispecific Zyboby, tetraspecific Zyboby, pentaspecific Zyboby, sextaspecific Zyboby, septaspecific Zyboby, octaspecific Zyboby, Knob-into-holes and duobodies.

The skilled person will know that there are multiple ways of constructing such multispecific antibodies or fragments thereof. For details on possible multispecific antibody formats, see
 20 Brinkmann, et al., MAbs, 2017, 9:182-212, Spiess, et al.; Molecular Immunology, 2015, 67:95-106; LaFleur, et al., mAbs, 2013, 5:208-218; US20170073415; Holliger and Hudson Nat. Biotechnol. 2005, 23:1126-1136; Kontermann, R.E., 2011, Bispecific Antibodies, Springer Science & Business Media. These are incorporated by reference in the present description.

In a preferred embodiment, the multispecific antibodies have a format selected from the group
 25 comprising trispecific triabodies, trispecific tribodies, IgG(H)-scFv, scFv-(H)IgG, IgG(L)-scFv, scFv-(L)IgG, 2scFv-IgG, IgG-2scFv, DVI-IgG, scFab-Fc(kih)-scFv2, scFab-Fc(kih)-scFv, IgG-taFv, scFv4-IgG and TVD-Ig.

Typically, an antibody or antigen-binding fragment of the invention which is modified in some way retains at least 10% of its original binding activity (when compared to the parental
 30 antibody) when that activity is expressed on a molar basis. Preferably, an antibody or antigen-binding fragment of the invention retains at least 20%, 50%, 70%, 80%, 90%, 95% or 100% or more of the binding affinity of the parental antibody. It is also intended that an antibody or antigen-binding fragment of the invention can include conservative or non-conservative amino acid substitutions (referred to as "conservative variants" or "function conserved variants" of the antibody) that do not
 35 substantially alter its biologic activity.

The present invention includes antibodies and antigen-binding fragments thereof and methods of use thereof. These antibodies or antigen-binding fragments thereof are at least partially free of other biological molecules from the cells or cell cultures in which they are produced and may be

considered "isolated". Such biological molecules include nucleic acids, proteins, lipids, carbohydrates, or other material such as cellular debris and growth medium. An isolated antibody or antigen-binding fragment may further be at least partially free of expression system components such as biological molecules from a host cell or of the growth medium thereof. Generally, the term "isolated" is not intended to refer to a complete absence of such biological molecules or to an absence of water, buffers, or salts or to components of a pharmaceutical formulation that includes the antibodies or fragments.

The present invention includes chimeric antibodies (*e.g.*, human constant domain/mouse variable domain) and methods of use thereof. As used herein, a "chimeric antibody" is an antibody having the variable domain from a first antibody and the constant domain from a second antibody, where the first and second antibodies are from different species. (U.S. Pat. No. 4,816,567; and Morrison et al., Proc. Natl. Acad. Sci. USA, 1984, 81: 6851-6855). Typically, the variable domains are obtained from an antibody from an experimental animal (the "parental antibody"), such as a rodent, and the constant domain sequences are obtained from human antibodies, so that the resulting chimeric antibody will be less likely to elicit an adverse immune response in a human subject than the parental (*e.g.*, mouse) antibody.

The present invention preferably includes humanized antibodies and antigen-binding fragments thereof (*e.g.*, rat or mouse antibodies that have been humanized) and methods of use thereof. As used herein, the term "humanized antibody" refers to forms of antibodies that contain sequences from both human and non-human (*e.g.*, mouse or rat) antibodies. In general, the humanized antibody will comprise substantially all of at least one, and typically two, variable domains, in which all or substantially all of the hypervariable loops correspond to those of a non-human immunoglobulin, and all or substantially all of the framework (FR) regions are those of a human immunoglobulin sequence. The humanized antibody may optionally comprise at least a portion of a human immunoglobulin constant region (Fc).

In general, the basic antibody structural unit comprises a tetramer. Each tetramer includes two different pairs of polypeptide chains, each pair having one "light" (about 25 kDa) and one "heavy" chain (about 50-70 kDa). The amino-terminal portion of each chain includes a variable region of about 100 to 110 or more amino acids primarily responsible for antigen recognition. The carboxy-terminal portion of the heavy chain may define a constant region primarily responsible for effector function. Typically, human light chains are classified as kappa and lambda light chains. Furthermore, human heavy chains are typically classified as mu, delta, gamma, alpha, or epsilon, and define the antibody's isotype as IgM, IgD, IgG, IgA, and IgE, respectively. Within light and heavy chains, the variable and constant regions are joined by a "J" region of about 12 or more amino acids, with the heavy chain also including a "D" region of about 10 more amino acids. See generally, Fundamental Immunology Ch. 7 (Paul, W., ed., 2nd ed. Raven Press, N.Y. (1989)).

The variable regions of each light/heavy chain pair form the antibody binding site.

Typically, the variable domains of both the heavy and light chains comprise three hypervariable regions, also called complementarity determining regions (CDRs), located within relatively conserved framework regions (FR). The CDRs are usually aligned by the framework regions, enabling binding to a specific epitope. In general, from N-terminal to C-terminal, both light and heavy chains variable domains comprise FR1, CDR1, FR2, CDR2, FR3, CDR3 and FR4. The assignment of amino acids to each domain is, generally, in accordance with the definitions of Sequences of Proteins of Immunological Interest, Kabat, et al.; National Institutes of Health, Bethesda, Md.; 5th ed.; NIH Publ. No. 91-3242 (1991); Kabat Adv. Prot. Chem. 1978, 32:1-75; Kabat, et al., J. Biol. Chem. 1977, 252:6609-6616; Chothia, et al., J Mol. Biol. 1987, 196:901-917 or Chothia, et al., Nature 1989, 342:878-883.

As used herein, the term "hypervariable region" refers to the amino acid residues of an antibody or antigen-binding fragment thereof that are responsible for antigen-binding. The hypervariable region comprises amino acid residues from a "complementarity determining region" or "CDR" (*i.e.* LCDR1, LCDR2 and LCDR3 in the light chain variable domain and HCDR1, HCDR2 and HCDR3 in the heavy chain variable domain). See Kabat *et al.* (1991) Sequences of Proteins of Immunological Interest, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md. (defining the CDR regions of an antibody by sequence); see also Chothia and Lesk J. Mol. Biol. 1987, 196: 901-917 (defining the CDR regions of an antibody by structure). As used herein, the term "framework" or "FR" residues refers to those variable domain residues other than the hypervariable region residues defined herein as CDR residues.

[0018] In this context, "sequence similarity" is based on the extent of identity combined with the extent of conservative changes. The percentage of "sequence similarity" is the percentage of amino acids or nucleotides which is either identical or conservatively changed viz. "sequence similarity" = percent sequence identity) + percent conservative changes). Thus, for the purpose of this invention "conservative changes" and "identity" are considered to be species of the broader term "similarity". Thus, whenever the term sequence "similarity" is used it embraces sequence "identity" and "conservative changes". According to certain embodiments the conservative changes are disregarded and the percent sequence similarity refers to percent sequence identity. In certain embodiments, the changes in a sequence permitted by the referenced percent sequence identity are all or nearly all conservative changes; that is, when a sequence is 90% identical, the remaining 10% are all or nearly all conservative changes. The term "nearly all" in this context refers to at least 75% of the permitted sequence changes are conservative changes, more preferably at least 85%, still more preferably at least 90%, and most preferably at least 95%. In certain embodiments of antibody heavy and/or light chains, the permitted sequence changes are within the framework regions and not in the CDRs.

[00192] The following references relate to BLAST algorithms often used for sequence analysis: BLAST ALGORITHMS: Camacho, C. et al. (2009); BMC Bioinformatics 10:421; Altschul et al. (2005) FEBS J. 272(20): 5101-5109; Altschul, S.F., et al., (1990) J. Mol. Biol. 215:403-410; Gish, W., et al., (1993) Nature Genet. 3:266-272; Madden, T.L., et al., (1996) Meth. Enzymol. 266:131-141; Altschul, S.F., et al., (1997) Nucleic Acids Res. 25:3389-3402; Zhang, J., et al., (1997) Genome Res.

7:649-656; Wootton, J.C., et al., (1993) *Comput. Chem.* 17:149-163; Hancock, J.M. et al., (1994) *Comput. Appl. Biosci.* 10:67-70; ALIGNMENT SCORING SYSTEMS: Dayhoff, M.O., et al., "A model of evolutionary change in proteins." in *Atlas of Protein Sequence and Structure*, (1978) vol. 5, suppl. 3. M.O. Dayhoff (ed.), pp. 345-352, Natl. Biomed. Res. Found., Washington, DC; Schwartz, R.M., et al.,
5 "Matrices for detecting distant relationships." in *Atlas of Protein Sequence and Structure*, (1978) vol. 5, suppl. 3." M.O. Dayhoff (ed.), pp. 353-358, Natl. Biomed. Res. Found., Washington, DC; Altschul, S.F., (1991) *J. Mol. Biol.* 219:555-565; States, D.J., et al., (1991) *Methods* 3:66-70; Henikoff, S., et al., (1992) *Proc. Natl. Acad. Sci. USA* 89:10915-10919; Altschul, S.F., et al., (1993) *J. Mol. Evol.* 36:290-300; ALIGNMENT STATISTICS: Karlin, S., et al., (1990) *Proc. Natl. Acad. Sci. USA* 87:2264-2268;
10 Karlin, S., et al., (1993) *Proc. Natl. Acad. Sci. USA* 90:5873-5877; Dembo, A., et al., (1994) *Ann. Prob.* 22:2022-2039; and Altschul, S.F. "Evaluating the statistical significance of multiple distinct local alignments." in *Theoretical and Computational Methods in Genome Research* (S. Suhai, ed.), (1997) pp. 1-14, Plenum, New York. In the present application, percent identity comparisons are preferably performed by a BLAST algorithm wherein the parameters of the algorithm are selected to give the
15 largest match between the respective sequences over the entire length of the respective reference sequences (e.g. expect threshold: 10; word size: 6; max matches in a query range: 0; BLOSUM 62 matrix; gap costs: existence 11, extension 1; conditional compositional score matrix adjustment).

In another embodiment, provided is a multispecific binding molecule or antigen-binding fragment thereof comprising at least a first binding site binding to a first target selected from the
20 group comprising human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, TDP43, mHTT, and fragments thereof and has V_L domains and V_H domains with at least 99% 98%, 97%, 96%, 95%, 90%, 85%, 80% or 75% sequence identity to one or
25 more of the V_L domains or V_H domains described herein and exhibits binding to the first target, and at least a second binding site binding to a second target selected from the group comprising α -synuclein, TDP43, mHTT, C1q, C5a, IL1B, IL6, TNF- α , APOE, IL12, IL23, human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and
30 aggregated human tau protein and cleaved and truncated versions thereof, and fragments thereof and has V_L domains and V_H domains with at least 99%, 98%, 97%, 96%, 95%, 90%, 85%, 80% or 75% sequence identity to one or more of the V_L domains or V_H domains described herein and exhibits binding to the second target, wherein the first and second target are different.

In another embodiment the multispecific binding molecule or fragment thereof of the
35 present invention comprises V_L and V_H domains (with and without signal sequence) as shown herein and additionally having up to 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 or more amino acid substitutions, while still exhibiting specific binding to the first and second target.

The skilled person will know that there are a number of monospecific antibodies known in the art that are able to bind to pathological proteins that are involved in NDDs targeted in the present invention. The CDRs of these antibodies are non-limiting examples of binding sites that can be employed in the multispecific binding molecules and are listed in Table 4. Additionally, there are also hybridomas known in the art that can produce monospecific antibodies, which can be employed in the multispecific binding molecules of the invention. For example, the C1q hybridomas 4A4B11, 23B6C8, 5B5C22 and 12A5B7 are particularly suitable for producing antibodies that can be employed in the multispecific binding molecules of the invention.

Table 4; Monospecific binding molecules against proteins involved in the pathology of NDD's

Target	name	binding SEQ.	Reference
Tau	BIB092	KYGMS (CDR-1 (HC)) TISSSGSRYYPSVKG (CDR-2 (HC)) SWDGAMDY (CDR-3 (HC)) KSSQSIVHSNGNTYLE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSLVPWA (CDR-3 (LC))	WO2015081085A2
Tau	C2N 8E12	EVKVVESGGGLVQPGGSMKLSVVSGFTFSNYWVNWVRQAP GKCLEWVAQIRLKSDNYATHYEESVKGRFTISRDDSKSSVYLQ MNNLRAEDSGIYYCTNWEDYWGGTTVTVSSASTKGPSVF (VH) DIVLTQSPDSLAVSLGERATISCRASQSVSTSRYSYIHWHYQQKPG QPPKLLIKYASNLESGVPSRFSGSGSGTDFTLNHPLEPEDFATY YCHHSWEIPLTFGQGTKLEIK (VL)	WO2015200806A2
Tau	RG7345		Roche RO6926496
Tau	RO 7105705 (37D3-H9)	SYGMS (CDR-1 (HC)) TINSGGTYYTPDSVKG (CDR-2 (HC)) SYSGAMDY (CDR-3 (HC)) RSSQSIVHSNGNTYFE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSLVPWT (CDR-3 (LC)) EVQVESGGD LARPGGSLKL SCTASGLIFE SYGMSVFPQT FQKLEWVAQIRLKSDNYATHYEESVKGRFTISRDDSKSSVYLQ MNNLRAEDSGIYYCTNWEDYWGGTTVTVSSASTKGPSVF (HC) DIVLTQSPDSLAVSLGERATISCRASQSVSTSRYSYIHWHYQQKPG QPPKLLIKYASNLESGVPSRFSGSGSGTDFTLNHPLEPEDFATY YCHHSWEIPLTFGQGTKLEIK (LC)	WO2016196726

Tau	54C1-H11	SYGMS (CDR-1 (HC)) TISSGGNYTYYPDSVKG (CDR-2 (HC)) SYSGAMDY (CDR-3 (HC)) RSSQSIVHSNGNTYLE (CDR-1 (LC)) TVSNRFS (CDR-2 (LC)) FQGSHPWT (CDR-3 (LC)) EVQLVESGGD LVKPGGSLKV SCVASGTFPH SYGMSWVRQT FDKRLDWAT ISSGGNYTYYPDSVKGFTI SRNAKNTLY LQMSLKSSED TAMYTCASSY SGAMDYWGQG TSVTVES (VH) QVMTQSPRLS LPVSLGDSAS ISCRSSQSIIV HSNGNTYLEW YLQKPGQSPK LLITPVSNRF SGVPDFTGGS GSGTDFTLKI SAVRAEDLEV YVCFQGSHPV WTEGGSTKLE IK (VL)	WO2016196726
Tau	123E9	DYYMK (CDR-1 (HC)) DIDPNNGGTSYNQKFKG (CDR-2 (HC)) SAGFEDSFEP (CDR-3 (HC)) RSSQSIVHSNGNTYLE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSHPPT (CDR-3 (LC)) EVQLQDSGPE LVKPGASVHM SCVASGTFPH DYYMKWVRQS KERSLEWIGD IDPNNGGTSY NQKFKGATL TVKRSSTAY MQLNLTRED SAVYYCARSA SPGDSFSTNG LQTLATVFA (HC) DVMTQSPRLS LPVSLGDSAS ISCRSSQSIIV HSNGNTYLEW YLQKPGQSPK LLITPVSNRF SGVPDFTGGS GSGTDFTLKI SAVRAEDLEV YVCFQGSHPV WTEGGSTKLE IK (LC)	WO2016196726
Tau	24A11	DYYMK (CDR-1 (HC)) DLNPKNGGII YNQKFKG (CDR-2 (HC)) SGGYGDSFAY (CDR-3 (HC)) RSSQIVHSN GNTYLE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSHPPT (CDR-3 (LC)) EVQLQDSGPE LVKPGASVHM SCVASGTFPH DYYMKWVRQS KERSLEWIGD IDPNNGGTSY NQKFKGATL TVKRSSTAY MQLNLTRED SAVYYCARSA SPGDSFSTNG LQTLATVFA (HC) DVMTQSPRLS LPVSLGDSAS ISCRSSQSIIV HSNGNTYLEW YLQKPGQSPK LLITPVSNRF SGVPDFTGGS GSGTDFTLKI SAVRAEDLEV YVCFQGSHPV WTEGGSTKLE IK (LC)	WO2016196726

Tau	Hu-37D3	SYGMS (CDR-1 (HC)) TINSGGTYTTYYPDSVKG (CDR-2 (HC)) SYSCANDY (CDR-3 (HC)) RSSQSIVHSNGNTYPE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGS LVPWT (CDR-3 (LC)) EVQLVESGGG LVQPSSSLRL SCASGLIPF SYGMSFVRA PKGLEWVAT INSGGTYTY PDSVKGFTI SHNSGNTLY LQMSLRAED TAVTYCANSY SCANDYFQGS TLVTYSS (HC) EDQLTQSPSS LSASVGDRTV ITCRSSQSIV HSNG YQOKPGKSPK LLIYKVSNRK SGVPSRFSGS GSGT SSLQPEDFAT YYCFQGS LVP WTFQGCTKVE IK (LC)	WO2016196726
Tau	Hu-125B11	NYWMN (CDR-1 (HC)) QIRLKSDNYATHYAESVKG (CDR-2 (HC)) GTTY (CDR-3 (HC)) KASQNVGTAVA (CDR-1 (LC)) SASIFYT (CDR-2 (LC)) QQFRTPYT (CDR-3 (LC)) EVQLVESGGG LVQPSSSLRL SCASGLIPF SYGMSFVRA PKGLEWVAT INSGGTYTY PDSVKGFTI SHNSGNTLY LQMSLRAED TAVTYCANSY SCANDYFQGS TLVTYSS (VH) DIQMTQSPSS LSASVGDRTV ITCRSSQNVG TAVNYDQEP GKSPFLLEYS ASIRYTGVPF PPSGSSSGTD PELTISSLQP EDENTYFCQQ PRTYPTFGQ QTNVRIK (VL)	WO2016196726

Tau	Hu-94B2	GYTMN (CDR-1 (HC)) LISPYNGVTSYNQKFKG (CDR-2 (HC)) QGRY (CDR-3 (HC)) KSSQSLLDSGKTYLN (CDR-1 (LC)) LVSKLDS (CDR-2 (LC)) WQGTHTFPWT (CDR-3 (LC)) EVQLVDSGAE VKKPGASVGV SOKASQYSIT GYTMNVPQA PQGLSEIGL ISPYNGVTSY SQKFSRATL TVLSTSTAY LRLSLLPSED TAVFYCARQG NYWQSTLYT VES (HC) DIYMTQTELS LSVTPGQFAS ISCRSSQSLL DSDGKTYLW LLQKPGQSPQ PLIYLVSKLD SGYPRFPGS GSSTDFTLKI SRVEAKGVGV YVCRQGYHTP STFGSTVE IK (LC)	WO2016196726
Tau	DC8E8	DYVIS (CDR-1 (HC)) EIFPRSGSTYYNEKFK (CDR-2 (HC)) DYYGTSFAMDY (CDR-3 (HC)) KSSQSLLNSRTRKNYL (CDR-1 (LC)) WASTRES (CDR-2 (LC)) KQSFYLRT (CDR-3 (LC))	WO 2016079597 A1
Tau	Tau13		Am J Pathol. 2017 Jun;187(6):1399-1412.
Tau	HT7		Am J Pathol. 2017 Jun;187(6):1399-1412.
Tau	BIB076		CN 105324394 A
Tau	5A6		Galasko D., 1997, J. neurochem, 68.1: 430- 433.
α -synuclein	PRX002/9E4		Mov Disord. 2017 Feb; 32(2): 211-218. US 8673593 B2
α -synuclein	BIB054		Neurimmune NI-202

α -synuclein	NI-202.3G12	DFHH (CDR-1 (HC)) WSNPQSGNSSSAQRFQG (CDR-2 (HC)) PHDGAGNYRFD (CDR-3 (HC)) SGDALPKHYAH (CDR-1 (LC)) KDTERPS (CDR-2 (LC)) QSADVSTYVV (CDR-3 (LC)) EVQLVQSGAEVKKPGASVRLSCRASGYNFIDFHHWVRQAPGE GLEWMGWSNPQSGNSSSAQRFQGRVTMTTDTSMSAAYMDLN WLTLDLTAVYYCTRPHDGAGNYRFDTWGQGLTVTVSS (HC) QSVLTQPPSVSVAPGQTARITCSGDALPKHYAHWYQQKPGQVP IVVIYKDTERPSGIPERFSGSTGTTVTLTISGVQAEDEAHYYCQ SADVSTYVVFGGGTKLTVL (LC)	WO 2010069603 A1
α -synuclein	NI-202.12F4	KAWMS (CDR-1 (HC)) RIKSTADGGTTSYAAPVEG (CDR-2 (HC)) AH (CDR-3 (HC)) SGEALPMQFAH (CDR-1 (LC)) KDSERPS (CDR-2 (LC)) QSPDSTNTYEV (CDR-3 (LC)) EVQLVQSGGGLVEPGSLRLSCAVSGFDFAKAWMSWVRQAPG QGLQWVARIKSTADGGTTSYAAPVEGRFIISRDRNMLYLQM NSLKTEDTAVYYCTSAHWGQGLTVTVSS (HC) QSVLTQPPSVSVSPGQTARITCSGEALPMQFAHWYQQRPGKAP VIVVYKDSERPSGVPERFSGSSGTTATLTITGVQAEDEADYYC QSPDSTNTYEVFGGGTKLTVL (LC)	WO 2010069603 A1

α-synuclein	NI-202.3D8	TYAIS (CDR-1 (HC)) IISNDGSRKYYADSVKG (CDR-2 (HC)) KRGTYASRCKAFDF (CDR-3 (HC)) RASQSIGWLA (CDR-1 (LC)) DASNLES (CDR-2 (LC)) QQYDNYWT (CDR-3 (LC)) RASHDISNYLA (CDR-1 (LC)) AASSLQS (CDR-2 (LC)) VQYRTYPLT (CDR-3 (LC)) EVQLVESGGGVVQGRSLRLSCAASGFTFSTYAIWVRQAPGK GLEWVAIISNDGSRKYYADSVKGFRFTISRDNSTLDLEMNLSR DEDTAVYYCAKKRGTYASRCKAFDFWGQGLTVTVSS (HC) DIQLTQSPSTLSASVGDRVTITCRASQSIGWLAWYQQKPGKAP KLLIYDASNLESGVPSRFSGSGSGTEFTLTISSLQPDDFATYYCQ QYDNYWTFGQGTKVEIK (LC) EIVMTQSPSSLSASIGDRVFTFCRASHDISNYLAWFRQQPGKAP KSLIYAASSLQSGVPSRFSASGSGTDFTLTISSLQPEDFATYYCV QYRTYPLTFGQGLTRLGIK (LC)	WO 2010069603 A1
α-synuclein	GM285	AASGFTFSRFTMT (CDR-1 (HC)) AISGSGGGTSYADSVK (CDR-2 (HC)) AKNWAPFDY (CDR-3 (HC)) RASQSVRSYLA (CDR-1 (LC)) GASSRAT (CDR-2 (LC)) QQYGSSPWT (CDR-3 (LC)) EVQLLESGGGLVQPGGSLRLSCAASGFTFSRFTMTWVRQAPGK GLEWVSAISGSGGGTSYADSVKGRLTVSRDNSKNTLYLQMNSL RAEDTAVYYCAAKNWAPFDYWGQGLTVTVSS (HC) EIVLTQSPGTLSPGERATLSCRASQSVRSYLAWYQQKPGQA PRLIYGASSRATGIPDRFSGSGTDFTLTISRLEPEDFAVYYC EEYGSSPWTFGQGTKVEIK (LC)	WO 2017009312 A1

α -synuclein	GM37	GFTFSSYAMT (CDR-1 (HC)) AIRSNGDRTDYADSVK (CDR-2 (HC)) AKNWAPFDS (CDR-3 (HC)) ASQSVSSSYLA (CDR-1 (LC)) GASSRAT (CDR-2 (LC)) QQYGSSPWT (CDR-3 (LC)) AIRSSGDRTDYADSVK (CRD-2 (HC)) AIRSQGDRTDYADSVK (CRD-2 (HC)) AIRSHGDRTDYADSVK (CRD-2 (HC)) EVQLLESQGGGLVQTGGSLRLSCAASGFTFSSYAMTWVRQAPGK GLEWVSAIRSNGDRTDYADSVKGRFTISRDNQNTLYLQMNSL RAEDTAVYYCAKNWAPFDSWGQGLTVTVSS (HC) EIVLTQSPGTLSPGERATLSCRASQSVSSSYLAWYQQKPGQA PRLLIYGASSRATGIPDRFSGSGSGTDFTLTISRLEPEDFAVYYCE EYGSSPWTFGQGTKVEIK (LC)	WO 2017009312 A1
α -synuclein	BA1	GFTFNTYAMN (CDR-1 (HC)) RIRTKSNDYATYYADSV (CDR-2 (HC)) VGYRPYAMDY (CDR-3 (HC)) RSSQNIVHSNGNTYLE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSHVPLT (CDR-3 (LC)) EVQLVETGGGLVQPKGSLKLSCATSGFTFNTYAMNNWVRQAP GKGLEWVARIRTKSNDYATYYADSVRITISRDDSQSMYLYLQMN NLKTEDTAMYYCVRVGYRPYAMDYWGQGSVTVSS (HC) DVLMTQTPLSLPVSLGDAQASISCRSSQNIVHSNGNTYLEWYLQK PGQSPILLIYKVSNRFSQVPDRFSGSGSGTDFTLKISRVEAEDLV YYCFQGSHVPLTFGAGTKLELK (LC)	US 8968734 B2
α -synuclein	BA2	GFTFSNYAMS (CDR-1 (HC)) TVTSGGSYTYYPDSV (CDR-2 (HC)) QNFGSRGWYFDV (CDR-3 (HC)) RSSQSIVNSNGNTYLE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSHVPLT (CDR-3 (LC)) EVQLVESGGDLVKPGGSLKFSCAASGFTFSNYAMSSWVRQTPD KRLEWVATVTSGGSYTYYPDSVGRFTISRDNKNTLYLQLSS LKSEDTAMYFCARQNFGSRGWYFDVWAGTTVTVSS (HC) DVLMTQTPLSLPVSLGDAQASISCRSSQSIVNSNGNTYLEWYLQK PGQSPKLLIYKVSNRFSQVPDRFSGSGSGTDFTLKISRVEAEDLG VYYCFQGSHVPLTFGAGTLELK (LC)	US 8968734 B2

α -synuclein	BA3	GFTFSSYAMS (CDR-1 (HC)) TISNGGSYTYYPDSV (CDR-2 (HC)) HSDYSGAWFAY (CDR-3 (HC)) SASSSVSYMY (CDR-1 (LC)) RTSNLAS (CDR-2 (LC)) QQYHSYPYT (CDR-3 (LC)) EVMLVESGGGLVKPGGSLKLSAASGFTFSSYAMSWVRQTPEK RLEWVATISNGGSYTYYPDSVRFTISRDNKNTLYLQMSSLRSE DTAMYYCARHSDYSGAWFAYWGQGTLLTVSA (HC) QIVLTQSPAIMSASPGEKVTISCSASSSVSYMYWYQKPGSSPKP WYRTSNLASGVPARFSGSGGTSYSLTISSMEAEDAATYYCQQ YHSYPYTFGGGTKLEIK (LC)	US 8968734 B2
α -synuclein	BA4	GDSFTSGYWN (CDR-1 (HC)) YIRYSGNTYYNPSL (CDR-2 (HC)) SYDYDRAWFAY (CDR-3 (HC)) RSSQSLVHSNGNTYLH (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) SQSTHVPWT (CDR-3 (LC)) EVQLQESGPGLVKPSQTLSTLTCSTVGDSFTSGYWNWIRKFFGN KLEYMGYIRYSGNTYYNPSLRISITRDTSKNQYYLQLISVTEDT ATFYCARSYDYDRAWFAYWQGALVTSA (HC) DVVMTQTPLSLPVSLGDQASISCRSSQSLVHSNGNTYLHWYLQ KPGQSPKLLIYKVSNRFSGVPDRFSGSGGTDFTLKISRVEAEDL GVYFCSQSTHVPWTFGGGTKLEIK (LC)	US 8968734 B2
α -synuclein		GFSLTSYGVH (CDR-1 (HC)) VIWRGGSTDYSAAP (CDR-2 (HC)) LLRSVGGFAD (CDR-3 (HC)) RSSQTIHNNNGNTYLE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSHPFT (CDR-3 (LC))	US 8968734 B2
α -synuclein		GFTFSNYAMS (CDR-1 (HC)) TISTGGSYTYYPDSV (CDR-2 (HC)) HSDYSGAWFAY (CDR-3 (HC)) SASSSVSYMY (CDR-1 (LC)) RTSNLAS (CDR-2 (LC)) QQFHSYPYT (CDR-3 (LC))	US 8968734 B2
α -synuclein		GFTFTDYMS (CDR-1 (HC)) FIRNKANGYTTEYSASV (CDR-2 (HC)) DYGNYAMDY (CDR-3 (HC)) KSSQSLLYSSNQKNYLA (CDR-1 (LC)) WASTRES (CDR-2 (LC)) QQYYSYPYT (CDR-3 (LC))	US 8968734 B2

α -synuclein	5C1	GYTFTNYWMN (CDR-1 (HC)) ATNPNGYTDYNQRFKD (CDR-2 (HC)) GGHLAY (CDR-3 (HC)) RSSQSLFHSGKNTYLH (CDR-1 (LC)) RVSNRES (CDR-2 (LC)) SQSAHVPWT (CDR-3 (LC)) QVQLQQSGAELAKPGTSVQMSCKASGYTFTNYWMNWIKARP GQGLEWIGATNPNGYTDYNQRFKDKAILTADKSSNTAYMHL SSLTSEDSAVYFCASGGHLAYWQGGTVVTVSA (HC) DVVMTQIPLYLSVSPGDQASISCRSSQSLFHSGKNTYLHWYLQK PGQSPKLLINRVSNRFGVDPDRFSGSGSGTDFTLKISGVEAEDLG VYFCQSQAHPVWTFGGGKLEIR (LC)	US 20140127131 A1
α -synuclein	9E4	NYGMS (CDR-1 (HC)) SISSGGGSTYYPDNVKG (CDR-2 (HC)) GGAGIDY (CDR-3 (HC)) KSIQTLLYSSNQKNYLA (CDR-1 (LC)) WASIRKS (CDR-2 (LC)) QQYYSYPLT (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCAASGFTFSNYGMSWVRQAPG KGLEWVASISSGGGSTYYPDNVKGKRFITSRDDAKNSLYLQMNS LRAEDTAVYYCSRGGAGIDYWGQGTLLTVSS (HC) DIQMTQSPSSLSASVGDRTVTHCKSIQTLLYSSNQKNYLAWFQQ KPGKAPKLLIYWASIRKSGVPSRFSGSGSGTDFTLTISSLQPEDF ATYYCQQYYSYPLTFGGGKLEIK (LC)	US 20130108546 A1
α -synuclein	Syn-211		Cell Reports 2014, 7, 2054–2065
α -synuclein	H3C		Clayton D.F., Neuron, 1995, 15.2: 361-72.
Amyloid β	Gantenerumab		RO4909832, RG1450
Amyloid β	Ponezumab		PF-04360365 LaPorte SL et al., J. Mol. Biol. 2012, 421:525-536
Amyloid β	Solanezumab		LY-2062430
Amyloid β	Donanemab		LY-3002813
Amyloid β	GSK933776		Andreasen N et al., PLoS One 2015: 10(3):e0098153

Amyloid β	BAN2401		Tucker S et al. J Alzheimers Dis. 2015;43(2):575-88
Amyloid β	KHK6640		
Amyloid β	Crenezumab	GFTFSSYGMS (CDR-1 (HC)) SINSNGGSTYYPDSVK (CDR-2 (HC)) GDY (CDR-3 (HC)) RSSQSLVYSNGDTYLH (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) SQSTHVPWT (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCAASGFTFSSYGMSWVRQAPGK GLELVASINSNGGSTYYPDSVKGRFTISRDNKNSLYLQMNSLR AEDTAVYYCASGDYWGQGTTVTVSS (HC) DIVMTQSPLSLPVTGPASISCRSSQSLVYSNGDTYLHWYLQK PGQSPQLLIYKVSNRFSGVPRFSGSGSGTDFTLKISRVEAEDVG VYYCSQSTHVPWTFGQGTKVEIK (LC)	WO 2015120233 A1; WO 2015120280 A1
Amyloid β	Adacanumab	SYGMH (CDR-1 (HC)) VIWFDGTKKYYTDSVK (CDR-2 (HC)) DRGIGARRGPYYMDV (CDR-3 (HC)) RASQSISSYLN (CDR-1 (LC)) AASSLQS (CDR-2 (LC)) QSYSTPLT (CDR-3 (LC)) QVQLVESGGGVVQPGSLRLSCAASGFAFSSYGMHWVRQAPG KGLEWVAVIWFDGTKKYYTDSVKGRFTISRDNKNTLYLQMN TLRAEDTAVYYCARDRGIGARRGPYYMDVWGKGTITVTVSS (HC) DIQMTQSPSSLSASVGDRVTHTCRASQSISSYLNWYQQKPGKAP KLLIYAASSLQSGVPSRFSGSGSGTDFTLTISLQPEDFATYYCQ QSYSTPLTFGGGGTKVEIKR (LC)	WO 2014089500 A1

Amyloid β	9TL	GYYTEAYYIH (CDR-1 (HC)) RIDPATGNTKYAPRLQD (CDR-2 (HC)) LYSLPVY (CDR-3 (HC)) KSSQSLLYSDAKTYLN (CDR-1 (LC)) QISRDP (CDR-2 (LC)) LQGTHTPV (CDR-3 (LC)) QVQLVQSGAEVKKPGASVKVSCKASGYTEAYYIHVVRQAPG QGLEWMGRIDPATGNTKYAPRLQDRVTMTTRDTSTSTVYMELS SLRSEDVAVYYCASLYSLPVYWGQGTITVTVSS (HC) DVVMTQSPLSLPVTLGQPASISCKSSQSLLYSDAKTYLNWFQQR PGQSPRRLIYQISRDPGVDRFSGSGSGTDFTLKISRVEAEDVGV VYYCLQGTHYPVLFQGQTRLEIKRT (LC)	WO 2006036291 A2
Amyloid β	3D6 (Bapineuzumab)	NYGMS (CDR-1 (HC)) SIRSGGRTYYSDNVKG (CDR-2 (HC)) YDHYSGSSDY (CDR-3 (HC)) KSSQSLLDSGKTYLN (CDR-1 (LC)) LVSKLDS (CDR-2 (LC)) WQGTHTPRT (CDR-3 (LC))	WO 2002088306 A2
Amyloid β		GFTFSSYAMS (CDR-1 (HC)) AINASGTRTYADSVKG (CDR-2 (HC)) GKGNTHKPYGYVRYFDV (CDR-3 (HC)) RASQSVSSSYLA (CDR-1 (LC)) GASSRAT (CDR-2 (LC)) LQIYNMPI (CDR-3 (LC)) QVELVESGGGLVQPGGSLRLSCAASGFTFSSYAMSWVRQAPGK GLEWVSAINASGTRTYADSVKGRFTISRDN SKNTLYLQMNSL RAEDTAVYYCARGKGNTHKPYGYVRYFDVWGQGTITVTVSS (HC) DIVLTQSPATLSLSPGERATLSCRASQSVSSSYLA WYQQKPGQA PRLLIYGASSRATGVPARFSGSGSGTDFTLTISLEPEDFATYYCL QIYNMPITFGQGTKEIKRT (LC)	WO 2014056816 A1
Amyloid structure	NPT088		Levenson JM et al., Alzheimer's & Dementia: Translational Res & Clin Interventions 2(3):141-155

Amyloid structures	g3p	AETVESCLAK PHTENSFTNV WKDDKTLDRY ANYEGCLWNA TGVVVCTGDE TQCYGTWVPI GLAIPENEGG GSEGGGSEGG GSEGGGKPP EYGDTPPGY TYINPLDGTY PPGTEQNPNAN PNPSLEESQP LNTEMFQNNR FRNRQGALTV YTGTVTQGTID PVKITYYQYTP VSSKAMYDAY WNGKFRDCAF HSGFNEDPFV CEYQGQSSDL PQPPVNAAGG SGGGSGGGSE GGGSEGGGSE GGGSEGGGSG GSGSGDFDY EKMANANKGA MTENADENAL QSDAKGKLDV VATDYGAAID GFIGDVSLA NGNGATGDFG GSNSQMAQVG DGDNSPLMNN FRQYLPSPQ SVECRPFVS AGKPYEFSID CDKINLFRGV FAFLLYVATF MYVESTFANI LRNKES	US20160009766 A1 US20140335016 A1
TDP-43	NI-205.3F10	SQAMS (CDR-1 (HC)) ALSRTGDTWYADSVRG (CDR-2 (HC)) NYYSSFGYNWAAFHI (CDR-3 (HC)) RASQDVNNNYLA (CDR-1 (LC)) GASRRAT (CDR-2 (LC)) QQYGGSPPYT (CDR-3 (LC)) EVQLLESGLDLVQPGGSLRLSCAASGFTFSQAMSWVRQAPGK GLEWVSALSRTGDTWYADSVRGRFTVSRDDSKNIFYLEMNSL RAEDTAVYYCAKNYYSSFGYNWAAFHIWGQGTMTVSS (HC) EIVLTQSPGTLISLSPGERATLSCRASQDVNNNYLAWYQQKPGQ APRLLIYGASRRATGVPDFSGRGSCTDFTLTINRLEPEDFAMY FCQQYGGSPPYTFGQGTKLEIK (LC)	WO2013061163A2
TDP-43	NI-205.51C1	DYWMH (CDR-1 (HC)) RINLDGSDTIYADSVKG (CDR-2 (HC)) SRKSV (CDR-3 (HC)) TGSNTDVGAYDYVS (CDR-1 (LC)) DVDVRPS (CDR-2 (LC)) SSYTKSGTLV (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRISCTTSGFISDYWMHWVRQAPGK GLTWVSRINLDGSDTIYADSVKGRFTISRDNKNTLYLQMNSL RVEDTAIYYCARSRKSVWGQGTMTVSS (HC) QSALTQPASVSGSPQSITISCTGSNTDVGAYDYVSWSQLPGK APKFVIFDVDRPSGISDRFSGSKSGNTASLTISGLQAEDEADYY CSSYTKSGTLVFGGGTKVTVV (LC)	WO2013061163A2

TDP-43	NI-205.21G2	SYTLH (CDR-1 (HC)) WINAAFINTKYSQKFQG (CDR-2 (HC)) RASGSNGLDV (CDR-3 (HC)) QASRDITNYLN (CDR-1 (LC)) DASYLET (CDR-2 (LC)) QYDSVPLT (CDR-3 (LC)) QVQLVQSGAEVKKPGASVKVCKTSGYSFTSYTLHWVRQAPG HRPEWMGWINAAFINTKYSQKFQGRITLTRDTSANIAYLELRSL TTEDTAVYYCARRASGSNGLDVWGQGTTVTVSS (HC) DIQMTQSPSSLSASVGDRTITTCQASRDITNYLNWYQKPKGAP KLLIYDASYLETGVPSTFSGSGSGTHFTLTISLQPDFATYYCQ QYDSVPLTFGGGTKVEIK (LC)	WO2013061163A2
TDP-43	NI-205.8A2	DHGMH (CDR-1 (HC)) VIWLDGSSRFYADSVEG (CDR-2 (HC)) DRVASEGTAFDV (CDR-3 (HC)) WASQNVNHLYV (CDR-1 (LC)) DTSVRAA (CDR-2 (LC)) QHRSDWT (CDR-3 (LC)) QVQLVESGGGVVQPGKSLRLSCAASGFTFRDHGMHWVRQAPG KGLEWVAVIWLDGSSRFYADSVEGRFTISRDNKNTLYLQLTSL RAEDTAIYYCARDRVASEGTAFDVWGQGTMTVTSS (HC) EIVLTQSPATLSLSPGERATLSCWASQNVNHLYLVWYQQRPGQA PRLLLYDTSVRAAGIPARFIGSGSGTHFTLTISLPEPDSAVYYCE HRSDWTFGGGTKVEIK (LC)	WO2013061163A2
TDP-43	NI-205.15F12	GYMH (CDR-1 (HC)) VINPNGGSTNYAQKFKG (CDR-2 (HC)) LPVNIEVLDL (CDR-3 (HC)) RSSQTVLFSSNDKNYLA (CDR-1 (LC)) WASVRAS (CDR-2 (LC)) QQSSTAPLT (CDR-3 (LC)) QVQLVQSGTAVKKPGASVKVCKASGFSGFYGMHWVRQAP GQGLEWMGVINPNGGSTNYAQKFKGRITMSADTPARSVSMEL GSLRSDDTAMYYCARLPVNIEVLDLWGQGTTLVTSS (HC) DIVMTQSPDSLAVSLGERATINCRSSQTVLFSSNDKNYLAWYQ QKPGQPPKLLIYWASVRASGVPRFSGSGSGTDFSLTINGLQAE DVAVYYCQSSSTAPLTFFGGGTKVEIK (LC)	WO2013061163A2

TDP-43	NI-205.113C4	NYMH (CDR-1 (HC)) INPSGGRTSYAQKFQG (CDR-2 (HC)) QRPSGYSYGPSESYGNPTDDAFDV (CDR-3 (HC)) GGNIGSRGVH (CDR-1 (LC)) DDSRPS (CDR-2 (LC)) QVWDNSSDHLVV (CDR-3 (LC)) QVQLVQSGAEVKKPGASVKVSCKASGYTFTNYYMHWVRQAP GQGLEWMGHNPSGGRTSYAQKFQGRASMTDSTSTVYMEVI SLRSEDVAVYYCARQRPSGYSYGPSESYGNPTDDAFDVWGQ GTTVTVSS (HC) SYVLTQPPSVSVAPGQTARITCGGNIGSRGVHWYQQRPGQAP VLVVYDDSDRPSGIPERFSGSNSGDTATLTISRVEVGDEADYYC QVWDNSSDHLVVFSGGKLTVL (LC)	WO2013061163A2
TDP-43	NI-205.25F3	NYVMY (CDR-1 (HC)) FISYDGSNKYYPDSVKG (CDR-2 (HC)) DTYQYDSSTYYPYFYYYGMDV (CDR-3 (HC)) IGSSDVGGYNYVS (CDR-1 (LC)) EVSNRPS (CDR-2 (LC)) SSFASSTSVT (CDR-3 (LC)) QVQLVESGGGVVQGRSLRLSCAASGFTFSNYVMYWVRQAPG KGLEWVAFISYDGSNKYYPDSVKGRFTISRDNMNTLTLMDS LRAEDTAVYYCARDTYQYDSSTYYPYFYYYGMDVWGQTTV TVSS (HC) QSALTQPASVSGSPGQSITISCTSSDVGGYNYVSWYQHPGK APKLMIEVSNRPSGVSSRFSGSKSGNTASLTISGLQSEADYY CSSFASSTSVTFGGKLTVL (LC)	WO2013061163A2
TDP-43	NI-205.87E7	SYAMS (CDR-1 (HC)) AISGGGDRYSADSVKG (CDR-2 (HC)) GGGGEMTAVTMDGTYYGMDV (CDR-3 (HC)) TGTSSNVGTYKFVS (CDR-1 (LC)) DVTKRPS (CDR-2 (LC)) CSYAGSYTYV (CDR-3 (LC)) EVQLLESGGGLVQPGGSLRLSCAASGFTFSYAMSWVRQAPGK GLEWVSAISGGGDRYSADSVKGRFTISRDNKNTLYLQINSLR VEDTAVYYCAQGGGEMTAVTMDGTYYGMDVWGQTTVT SS (HC) QSALTQPRSVSGSPGQSITISCTGTSSNVGTYKFVSWYQHPGK APKLMIDVTKRPSGVDPDRFSGSKSGNTASLTISGLQAEADY YCCSYAGSYTYVFGSGTKVTVL (LC)	WO2013061163A2

TDP-43	NI-205.21G1	SHGMH (CDR-1 (HC)) VISYDASNKSYADSVKG (CDR-2 (HC)) AFSSSASGGY (CDR-3 (HC)) RSSQSLVHSDGVTYLN (CDR-1 (LC)) KVSNRDS (CDR-2 (LC)) MQGTHWPPWT (CDR-3 (LC)) QVQLVESGGGVVQPGMSLRSLSCAASGFSFSSHGHWVRQTPG KGLEWLAVISYDASNKSYADSVKGRFTISRDNSSKKTLYLQMDS LRVEDTALYYCANAFSSSASGGYWGQGTLLTVSS (HC) DVVMTQSPLSLPVTLGQPASISCRSSQSLVHSDGVTYLWNFQQR PGQSPRRLIYKVSNRDSGVPDRFSGSGSGTDFTLEISRVEAEDVG IYYCMQGTHWPPWTFGQGTKVEIK (LC)	WO2013061163A2
TDP-43	NI-205.68G5	SYGMH (CDR-1 (HC)) IIYYDSSQRYYADSVKG (CDR-2 (HC)) DLPFHYHRSASFAPSDT (CDR-3 (HC)) RASQAVTNNYLA (CDR-1 (LC)) AASSRAT (CDR-2 (LC)) QQYGTSPIT (CDR-3 (LC)) QVQLVESGGGVVQGRSLRLSCAASGFTFSSYGMHWVRQAPG KGLEWVAIIYYDSSQRYYADSVKGRFTISRDNSSKNALYLQMNS LRAEDTALYYCARDLPFHYHRSASFAPSDTWGQGTLLTVSS (HC) EIVLTQSPGTLSPGERATLSCRASQAVTNNYLAWYQQKPGQ APRLLVYAASSRATGIPDRFYSGSGADFTLTISRLEPEDFAVYY CQQYGTSPITFGQGTREIK (LC)	WO2013061163A2
TDP-43	NI-205.20A1	SYRMN (CDR-1 (HC)) YISTSSSTIYYADSVKG (CDR-2 (HC)) AFDY (CDR-3 (HC)) RASQSVSSSYLA (CDR-1 (LC)) GASSRAT (CDR-2 (LC)) QQYGSSPFT (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCAASGFTFSSYRMNWVRQAPG KGLEWVSYISTSSSTIYYADSVKGRFTISRDNKNSLYLQMNSL RAEDTAVYYCARAFDYWGQGTLLTVSS (HC) EIVLTQSPGTLSPGERATLSCRASQSVSSSYLAWYQQKPGQA PRLLIYGASSRATGIPDRFSGSGSGTDFTLTIRLEPEDFAVYYCQ QYGSSPFTFGQGTKVEIK (LC)	WO2013061163A2

mHTT	NI-302.33C11	DFGMH (CDR-1 (HC)) LIWYDGGYKYYADSVKG (CDR-2 (HC)) HLEYCSRTTCYLGH (CDR-3 (HC)) RASQGSDYLA (CDR-1 (LC)) AASTLQT (CDR-2 (LC)) QQLKTYPYT (CDR-3 (LC)) EVQLVESGGGVVQPGNSLRSLSCAASGFRFSDFGMHWVRQAPG KGLEWLALIWYDGGYKYYADSVKGRFTISRDN SKNTMFLQMN SLRAQDTAVYYCATHLEYCSRTTCYLGHWGQGTLLTVSS (HC) DIQLTQSPSFLSASVGDVTFTCRASQLAWFQKPGIAPKLLIYA ASTLQTGVPSRFGSGSGTEFTLTIRSLQSEDFGTYCQQLKTYP YTFGQGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.63F3	TRSMN (CDR-1 (HC)) WINTNTGNRTYVQAFRG (CDR-2 (HC)) GAGGGYWFD (CDR-3 (HC)) KSNQSLFYSSNNNNYLA (CDR-1 (LC)) WGSTRES (CDR-2 (LC)) HQYYHNPYT (CDR-3 (LC)) QVQLVQSGSSVKVCKASGYTFETRSMNWVRQAPGQGLEVMG WINTNTGNRTYVQAFRGFRVFSLDTSVSTAYLQISNLKTEDTAV YYCARGAGGGYWFD SWGQGTLLTVSS (HC) DIQMTQSPDSLAVSLGERATINCKSNQSLFYSSNNNNYLAWYQ HKSGQPPKLLVYWGSTRESGVPDRFSGSGSGTDFTLTISLQAE DVAIYYCHQYYHNPYTTFGQGTLEIK (LC)	WO2016016278A3
mHTT	NI-302.35C1	ITALS (CDR-1 (HC)) AITGNAYGTYADSVKG (CDR-2 (HC)) GIASDSSGYSF (CDR-3 (HC)) RASQSVDNQFA (CDR-1 (LC)) DASRRAP (CDR-2 (LC)) QHRYTWLYT (CDR-3 (LC)) EVQLVESGGNLVQPGGSLRLSCTASGFTFSITALSWVRQAPKPG PQWVSAITGNAYGTYADSVKGRFTISRDN AKNTLYLQMNGL RAEDTAIYYCVKGIASDSSGYSF WGPGLTVTVSS (HC) EIVLTQSPATLSLSPGERATLSCRASQSVDNQFAWYQKPGQAP RLLIYDASRRAPGIPDRFSGSGSGTDFTLTISLQEPEDFAIYYCQH RYTWLYTFGQGTLEIK (LC)	WO2016016278A3

mHTT	NI-302.31F11	STYMS (CDR-1 (HC)) VIFSGADTYADSVKG (CDR-2 (HC)) HYYGSDLPSDF (CDR-3 (HC)) RSSQSLLYSNGYNYLD (CDR-1 (LC)) LGSDRAS (CDR-2 (LC)) MQGLQSPWT (CDR-3 (LC)) EVQLVESGGGLIQPGGSLRLSCAASGFTVSSSTYMSWVRQAPGK GLQCVSVIFSGADTYADSVKGRFTVSRDNSKNTLFLQMNSLR VEDTATYYCVRHYYGSDLPSDFWGQGTLLTVSS (HC) DVVMTQSPLSLPVAPGEPASISCRSSQSLLYSNGYNYLDWYLQ KPGKPPQLLVYLGSDRASGVPDRFSGSGSGKDFTLNISRVEAED VGVYYCMQGLQSPWTFGQGTKLEIK (LC)	WO2016016278A3
mHTT	NI-302.2A2	TYWMN (CDR-1 (HC)) NIKPDGSDKYYVDSVKG (CDR-2 (HC)) GDGSGWNVY (CDR-3 (HC)) KSSQSLLYTSKNKDSKNYLG (CDR-1 (LC)) WASTRES (CDR-2 (LC)) QYYTTPQ (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCAASGFTFSTYWMNWVRQAPG KGLEWVANIKPDGSDKYYVDSVKGFTISRDNKNSLYLQMN SLRDEDTAVYYCARGDGSGWNVYWGQGTLLTVSS (HC) DIQMTQSPNSLAIVSLGERATINCKSSQSLLYTSKNKDSKNYLG WYQQKPGQPPKLLIYWASTRESGVPDRFSGSGSGTDFTLTISL QAEDVAVYYCQYYTTPQFGGGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.6N9	SYAMT (CDR-1 (HC)) TISATGGSTFYTDSVRG (CDR-2 (HC)) DLFGVDTSYYGMDV (CDR-3 (HC)) RPSQSVSGRYVA (CDR-1 (LC)) AASNRAI (CDR-2 (LC)) QHYGASSYT (CDR-3 (LC)) EVQLVESGGDLVQPGGSLRLSCVVSIGFTFSSYAMTWVRQAPGK GLAWVSTISATGGSTFYTDSVRGFTISRDNKNTLYLQMNSLR TDDTAIYYCVKDLFGVDTSYYGMDVWGQGTTVTVSS (HC) EIVLTQSPGTLSPGERATLSCRPSQSVSGRYVAWYQQKPGQA PRLIFYAASNRAIGIPDRFSGSGSGTDFTLTISRLEPEDFAVYYC QHYGASSYTFGPGTKVDIK (LC)	WO2016016278A3

mHTT	NI-302.74C11	GYFLH (CDR-1 (HC)) WINPNSGDTNYAE (CDR-2 (HC)) EAPDPGAETDV (CDR-3 (HC)) SGDAVPKQYIY (CDR-1 (LC)) KDTQRPS (CDR-2 (LC)) QSADSSATWV (CDR-3 (LC)) EVQLVQSGTEVQKPGASVKVSKASGYSTGYFLHWVRQAPG QGLEWMGWINPNSGDTNYAEKFRGRIMTRDTSVSTAHEMELSS LRFDDTALYYCTREAPDPGAETDVWGQGTITVTVSS (HC) QSVLTQPPSVSVSPGQTARITCSGDAVPKQYIYWYQQKPGQAPI LVIYKDTQRPSGIPERFSGSNSGTTVTLTITGVQADDEGDYYCQS ADSSATWVFGGGTKLTVL (LC)	WO2016016278A3
mHTT	NI-302.15F9	NSSNM (CDR-1 (HC)) SIDTSATNYKYYADSVKG (CDR-2 (HC)) GYTTPRDFDY (CDR-3 (HC)) RSSQSLLYRDNNTYLN (CDR-1 (LC)) RASDRDS (CDR-2 (LC)) MQGTHWPRT (CDR-3 (LC)) EVQLVESGGGLVTPGGSRLSCEASGFLFKNSSMNWVRQTPGK GLEWVSSIDTSATNYKYYADSVKGRFTISRDDATNSLYLQMNS LRADDTATYYCARGYYTPRDFDYWGQGTITVTVSS (HC) DVVMTQSPQTLVSLSGQAASISCRSSQSLLYRDNNTYLNWFHQ RPGQSPRRLIYRASDRDSGVPDRFSGGGSGTDFTLKISGVEAED VGTYYCMQGTHWPRTFGQGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.39G12	NYAIT (CDR-1 (HC)) VIYRDGRYYGDSVRG (CDR-2 (HC)) AHGQYYYGVDV (CDR-3 (HC)) RSSQSLHSNGYNYLD (CDR-1 (LC)) LSSNRPS (CDR-2 (LC)) MQSLQTFT (CDR-3 (LC)) EVQLVQSGGGLVHPWGSRLSCLASGFSVSNYAITWVRAPGK GLQYISVIYRDGRYYGDSVRGRFTISRDDSKNTLYLQMNSLRF EDTAVYYCARAHGQYYYGVGVWGQGTITVTVSS (HC) DVVMTQSPQLSVSPGEPASISCRSSQSLHSNGYNYLDWYRQK PGQSPQLLIYSSNRPSGVPDRFSASGSGTEFTLQISRVEAEDVG VYYCMQSLQTFTFGGGTKVDIK (LC)	WO2016016278A3

mHTT	NI-302.11A4	SSYMS (CDR-1 (HC)) VLYRDGDTYYADSVQG (CDR-2 (HC)) DRRSSHYYYGMDV (CDR-3 (HC)) RASQSVSSSYFA (CDR-1 (LC)) GTSRRAT (CDR-2 (LC)) QQYGSSWT (CDR-3 (LC)) EVQLVESGGGLIQPGGSLRLSCAASGFPVSSSYMSWVRQAPGE GLEWVSVLYRDGDTYYADSVQGRFTISRDNQNTFYLMNSL KAEDTAVYYCAGDRSSHYYYGMDVWGQGTITVTVSS (HC) EIVMTQSPGTLSPGERATLSCRASQSVSSSYFAWYQQKPGQA PRLLIYGTSRRATAIPDRFSGSGSGTDFTLTISRLEPEDFAVYYCQ QYGSSWTFGPGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.22H9	NYAIT (CDR-1 (HC)) VIYRDGRYYGDSVRG (CDR-2 (HC)) AHGQYYYGVDV (CDR-3 (HC)) RSSQSLHNSGYNLYD (CDR-1 (LC)) LNSNRAS (CDR-2 (LC)) MQSLQTFT (CDR-3 (LC)) EVQLVESGGGLVHPWGSLRVSCAASGFSVSNYAITWVRQAPG KLEYISVIYRDGRYYGDSVRGRFTISRDDSKNTIYLMNSLR FEDTAVYYCARAHGQYYYGVDVWGQGTITVTVSS (HC) DVVMTQSPLSLVSPGEPASISCRSSQSLHNSGYNLYDWYRQK PGQSPQLLIYLNNSNRASGVPDRFSGSGSGTEFTLTISRVEAEDVG VYYCMQSLQTFTFGGGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.44D7	SYAMS (CDR-1 (HC)) GIGYSDTSTYYADSVKG (CDR-2 (HC)) GTRDYYGMDV (CDR-3 (HC)) GLSSGSVSTSYPS (CDR-1 (LC)) STNTRSS (CDR-2 (LC)) VLFMSGIGV (CDR-3 (LC)) EVQLVQSGGGLVQPGGSLRLSCAASGFTFSSYAMSWVRQAPG KLEWVSGIGYSDTSTYYADSVKGRFTVSRDISKNTLYLMNS LRAEDTAVYYCAKGTRDYYGMDVWGQGTITVTVSS (HC) QTVVTQEPSFSVSPGGTVTLTCGLSSGSVSTSYPSWYQQTPGR APRTLIYSTNTRSSGVPDRFSGSILGNKAALTITGAQADDESYY CVLFMSGIGVFGGGTRLTVL (LC)	WO2016016278A3

mHTT	NI-302.37C12	NSQMT (CDR-1 (HC)) VIYTSGSAYYADSVK (CDR-2 (HC)) GPSAYYYGLDL (CDR-3 (HC)) RSSQSLHSNGYNYLD (CDR-1 (LC)) LGSTRAS (CDR-2 (LC)) MQGLQTYT (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCVASALTVTNSQMTWVRRAPG RGLEWVSVIYTSGSAYYADSVKGRFTISRDNKNTVFLQMNSL RVEDTAVYYCAKGPSAYYYGLDLWGQGTTVTVSS (HC) DIVMTQSPLSLPVTGPGEASISCRSSQSLHSNGYNYLDWYLQK PGQSPQLLIYLGSTRASGVPDFRSGSGSGTDFTLKISRVEAEDVG VYYCMQGLQTYTFGQGTKLEIK (LC)	WO2016016278A3
mHTT	NI-302.55D8	DYYIH (CDR-1 (HC)) RINPNNGGTNYAQNFGQWVTMT (CDR-2 (HC)) VGSELLREGGYHYMDV (CDR-3 (HC)) TGSSNIGAGYDVH (CDR-1 (LC)) DNTNRPS (CDR-2 (LC)) QSYDNSLSGSWV (CDR-3 (LC)) QVQLVQSGSEVKKPGASVKVSKASGYTFTDYYIHWVRQAPG QGLEWMGRINPNNGGTNYAQNFGQWVTMTDRDTSISTAYMELS RLRSDDTAVYYCARVGSELLREGGYHYMDVWGKGTITVS S (HC) QSVLTQPPSVSGAPGQRVTISCTGNSSNIGAGYDVHWYQQLPG TAPKLLIFDNTNRPSGVPDFRSGSKSGTSASLAITGLQAEDEANY HCQSYDNSLSGSWVFGGKTLTVL (LC)	WO2016016278A3
mHTT	NI-302.7A8	NSWMT (CDR-1 (HC)) NIKEDGSRTYYVDSVKG (CDR-2 (HC)) GDYNSGIYYFPGDY (CDR-3 (HC)) RSSQSLVYSDGNTYLN (CDR-1 (LC)) KVSNRDS (CDR-2 (LC)) MQGTHWPGT (CDR-3 (LC)) EVQLVESGGGSVQPGGSLRLSCVASGFIFRNSWMTWVRQDPGK GLEWVANIKEDGSRTYYVDSVKGGRFTISRDNKNSLYLQMNSL RAEDTAVYYCARGDYNSGIYYFPGDYWGQGTLVTVSS (HC) DVVMQSPSLPVTLGQPASISCRSSQSLVYSDGNTYLNWFQQR PGQSPRLIYKVSNRDSGVPDFRSGSGSGTDFTLRISRVEAEDVG IYYCMQGTHWPGTFGQGTKVEIK (LC)	WO2016016278A3

mHTT	NI-302.78H12	NGYYWG (CDR-1 (HC)) SIYHNGNTYYNPSLKS (CDR-2 (HC)) PSATYYYGSGTQFHAFDV (CDR-3 (HC)) TGTSRDVGNYNYVS (CDR-1 (LC)) DVSERPS (CDR-2 (LC)) CSYAGSYTFEV (CDR-3 (LC)) QVQLQESGPGLVKPSSETLSLTCLVSSYSISNGYYWGWRQPPGK GLEWIGSIYHNGNTYYNPSLKSRIISVDTSKNQFSLKLRSVTAA DTAVYYCAMP SATYYYGSGTQFHAFDVWGQGTITVTVSS (HC) QSALTQPRSVSGSPGQSVTISCTGTSRDVGNYNYVSWYQQHPG EVPKLIYDVSRPSGVPDRFSGSKSGNTASLTISGLQAED EADY YCCSYAGSYTFEVFGGGTKLTVL (LC)	WO2016016278A3
mHTT	NI-302.71F6	GYYSWS (CDR-1 (HC)) EVNHSGGTNLNSSLKS (CDR-2 (HC)) GYSYDPKYYFDS (CDR-3 (HC)) TGTSSDIGSYDFVS (CDR-1 (LC)) GVNKRPS (CDR-2 (LC)) CSYAGSTTWV (CDR-3 (LC)) QVQLQQWGAGLLKPSSETLSLTCAVYGGSLSGYYWSWRQPPG KLEWIGEVNHSGGTNLNSSLKSRIISVDKSKKQFSLKLSVT AADTAMYFCARGYSYDPKYYFDSWSQGTITVTVSS (HC) QSALTQPASVSGSPGQAITISCTGTSSDIGSYDFVSWYQQDPGK APKVIIYGVNKRPSGVSNRFGSKSGNTASLTISGLQAD EADY YCCSYAGSTTWVFGGGTKLTVL (LC)	WO2016016278A3
mHTT	NI-302.11H6	TYSFT (CDR-1 (HC)) WISAYNGHTNYVDSFQG (CDR-2 (HC)) VDTTYYYYGMDV (CDR-3 (HC)) ALRFGSVSSSYPS (CDR-1 (LC)) STNTRSS (CDR-2 (LC)) VLYMGSGIGV (CDR-3 (LC)) EVQLVQSGAVMKKPGDSVRVSCRASTYSFSTYSFTWVRQVPG QGLEWMGWISAYNGHTNYVDSFQGRLLTDTASTAYMELR SLRSDDTAIYYCAAVDTTYYYYGMDVWGQGTITVTVSS (HC) QTVVTQEPTFSVSPGGTVTLTALRFGSVSSSYPSWFQQTPGQ APRTLIIYSTNTRSSGVPARFSGSILGNKAALTIAGAQANDEADY YCVLYMGSGIGVFGGGTKLTVL (LC)	WO2016016278A3

mHTT	NI-302.3D8	TYAMS (CDR-1 (HC)) AISATGGSTFYAESVKG (CDR-2 (HC)) GSTAVYLFDS (CDR-3 (HC)) RASQDIRNFLAW (CDR-1 (LC)) AASTLQS (CDR-2 (LC)) QQFYNYPPPT (CDR-3 (LC)) EVQLVQSGGGLVQPGGSLRLSCEASGFIFKTYAMSWVRQLPGR GLEWVSAISATGGSTFYAESVKGRLTISRDTAKNTVYLQMNLL RAEDTAMYYCAKGSTAVYLFDSWGQGLTVTVSS (HC) DIQMTQSPSSLSASVGDRTLTCSRASQDIRNFLAWIQKPGKPP KSLIYAASTLQSGVPSRFSGSGSGTDFTLTISSLHPEDFATYYCQ QFYNYPPPTFGQGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.18A1	TDYYY (CDR-1 (HC)) TIYFGGATYYNPSLRN (CDR-2 (HC)) VGYLDRSGLL (CDR-3 (HC)) RSSQSLHNNGYNYLD (CDR-1 (LC)) LGSTRAS (CDR-2 (LC)) MQALQTPPT (CDR-3 (LC)) QLQLQESGPGLVKPSEALSLCTVSGGSITTDYYYWGWRQSPG KGLEWVGTIYFGGATYYNPSLRNRVSVSDTSNTRLRLISLSA ADTAVYYCARVGYLDRSGLLVGQGLTVTVSS (HC) EIVLTQSPLSVPVTPGEPASISCRSSQSLHNNGYNYLDWYLLKP GQSPQLLIYLGSTRASGVPDFSASGSGTDFTLEISRVEAEDVGV YYCMQALQTPPTFGRTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.8F1	NAWMN (CDR-1 (HC)) HIRTQAEGGTSDYAAPVKG (CDR-2 (HC)) PPYYYYYGLDV (CDR-3 (HC)) TGASSDVGTVDLVS (CDR-1 (LC)) EVNKRPS (CDR-2 (LC)) CSYAGYSTV (CDR-3 (LC)) EVQLVESGGGLVQPGGSLTISCAASGFTFSNAWMNWVRQAPG KGLEWVGHIRTQAEGGTSDYAAPVKGRFTISRDDSKNTLYLQM NSLKTEDTAVYYCIPPPYYYYYGLDVWGQGTITVTVSS (HC) QSALTQPASVSGSPGQSITISCTGASSDVGTVDLVSQYQHPGK APKLIIEVNKRPSGVSYRFSASKSANTASLTISGLQAEDEAEYY CCSYAGYSTVFGGGTKLTVL (LC)	WO2016016278A3

mHTT	NI-302.52C9	DTYMS (CDR-1 (HC)) GIHAGGETYYADSVKG (CDR-2 (HC)) HYYGNDDDDTDY (CDR-3 (HC)) RSSQSLHSNGYNYLD (CDR-1 (LC)) LGSTRAS (CDR-2 (LC)) LQAQQIPWT (CDR-3 (LC)) EVQLVQSGGGLVQPGGSLRLSCAGSGFTVSDTYMSWVRQAPG KGLEWVSGIHAGGETYYADSVKGRFTISRDN SKNTLYLQMNRL TPEDTAVFYCARHYYGNDDDDTDYWGQGT LVTVSS (HC) DVVM TQSPLSLPVTGPGEPAISCRSSQSLHSNGYNYLDWYVQ KPGQSPQLLIYLGSTRASGVPDRFSGSGSGTDFTLKISRVEAEDV GVYYCLQAQQIPWTFGQG TKVEIK (LC)	WO2016016278A3
mHTT	NI-302.46C9	SGAYY (CDR-1 (HC)) RVYPTWSTNYNPSLES (CDR-2 (HC)) EAPGDYDAAPLAY (CDR-3 (HC)) RASQYISHYLN (CDR-1 (LC)) AASSLQS (CDR-2 (LC)) QQSYTPRT (CDR-3 (LC)) QVQLQESGPGLVKPSQTLSTCTVSGASVSSGAYYWSWIRQPA GKRLEWIGRVYPTWSTNYNPSLESRTISLDTSNQFSLKLTSL TAADTAVYYCAREAPGDYDAAPLAYWGQGT LVTVSS (HC) DIQMTQSPSSLSASVGDRTITCRASQYISHYLNWYRQKPGKAP QLVIYAASSLQSEVPSRFSGSGSGPEFTLTISLQPEDFATYYCQQ SYTTPRTFGQGT KLEIK (LC)	WO2016016278A3
mHTT	NI-302.64E5	QAWMS (CDR-1 (HC)) RIKTKTEGEATDYAAPVRG (CDR-2 (HC)) TGVLA AAVDVY (CDR-3 (HC)) KSSQSLFYSYNNENYLA (CDR-1 (LC)) WASTRES (CDR-2 (LC)) QQYYSTPQT (CDR-3 (LC)) EVQLVETGGGLVKPGGSLRLSCAASGFTFDQAWMSWVRQVPG KGLEWVGRIKTKTEGEATDYAAPVRGRFTISRDDSEDTVFLQM NSLKTEDTALYYCTSTGVLA AAVDVYWGQGT LVTVSS (HC) DIQLTQSPDSLAVSLGERATMTCKSSQSLFYSYNNENYLA WYQ QRPGQPPKLLIYWASTRESGVPDRFSGSGSGTDFLTISLQAEED VAVYYCQQYYSTPQTFGQGT KVDIK (LC)	WO2016016278A3

mHTT	NI-302.7D8	NYAIN (CDR-1 (HC)) WINTITGHPTYAQGFKG (CDR-2 (HC)) TYSNYGEFDY (CDR-3 (HC)) TGTSSDVGSYNLVS (CDR-1 (LC)) EGSERPS (CDR-2 (LC)) CSYAGTTTFVL (CDR-3 (LC)) QVQLVQSGSELKKPGASVKVSKASGYNFNYYAINWLRQAPG QGLEWMGWINTITGHPTYAQGFKGRFVFLDTSVSTAYLQISSL KPEDTAVYYCARTYSNYGEFDYWGQGLTVTVSS (HC) QSALTQPASVSGSRGQSITISCTGTSSDVGSYNLVSQYQYPGK APKLIIEGSRPSGVSNRFGSKSGNTASLTISGLQAEDEADYY CCSYAGTTTFVLFGGGTKLTVL (LC)	WO2016016278A3
mHTT	NI-302.72F10	SYAMS (CDR-1 (HC)) DISGIGSNTYYADSVKG (CDR-2 (HC)) DRKRSGWYEQ (CDR-3 (HC)) RASQSIAYLG (CDR-1 (LC)) DASIRAT (CDR-2 (LC)) HQRSKWPLT (CDR-3 (LC)) EVQLVETGGGFVQPGGSLRLSCAASGFNFGSYAMSWVRQAPG KGLEWVSDISGIGSNTYYADSVKGRFTISRDNSTLYLDMSSL RAEDTARYYCAKDRKRSGWYEQWGQGLTVTVSS (HC) EIVMTQSPATLTLSPPERATLSCRASQSIAYLGWYQKPGQAP RLLIYDASIRATGIPDRFSGSGSGTDFTLTISSEPEDSAVYYCHQ RSKWPLTFGGGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.4A6	AYAMS (CDR-1 (HC)) TISGSGSTYYADSVKG (CDR-2 (HC)) VTTELYGANSYYYYMDV (CDR-3 (HC)) RASQSVVSRYLA (CDR-1 (LC)) GASSRAT (CDR-2 (LC)) QLYGNSQT (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCAASGFTFSAYAMSWVRQAPG KGLEWVSTISGSGSTYYADSVKGRFSISRDNSTLYLQMNSL RAEDTAVYFCAKVTTELYGANSYYYYMDVWGKGTITVTVSS (HC) EIVLTQSPGTLTSLPPERATLSCRASQSVVSRYLAWYQKPGQA PRLIYGASSRATGIPDRFSGSGSGTDFTLTISRLEPEDFAMYYC QLYGNSQTFGQGTKVEIK (LC)	WO2016016278A3

mHTT	NI-302.12H2	NYAMG (CDR-1 (HC)) VISGTGGSTYYADSVKG (CDR-2 (HC)) DLRKISGPLYYYGMDV (CDR-3 (HC)) RASQSVSSGYLA (CDR-1 (LC)) GASTRAT (CDR-2 (LC)) QHYGASSYT (CDR-3 (LC)) EVQLVQSGGGLVQPGGSLRLSCEASGFTFSNYAMGWVRQAPG KGLEWVSVISGTGGSTYYADSVKGRFTISRDN\$MNTLYLQMNS PRADDTAVYYCAKDLRKISGPLYYYGMDVWGQGT\$TVTVSS (HC) EIVLTQSPGTL\$SLSPGERATL\$CRASQSVSSGYLA\$WYQQKPGQA PRLIYGA\$TRATGIPDRFSGSGSGTDFTLTISRLEPEDFAVYYCQ HYGASSYTFGQGTKLEIK (LC)	WO2016016278A3
mHTT	NI-302.8M1	IYYMH (CDR-1 (HC)) GISPSGAHTMYAQNFQG (CDR-2 (HC)) GSTVTNYRPF\$DY (CDR-3 (HC)) RASQDISNYLA (CDR-1 (LC)) AASTLQS (CDR-2 (LC)) QNYNSGPPP (CDR-3 (LC)) EVQLVQSGAEVKKPGASVKVSCKASGYTFTIYYMHWVRQAPG QGLEWMGGISPSGAHTMYAQNFQGRVTVTRDTSTSTVYMELS SLRSEDTAVYYCARGSTVTNYRPF\$YWGQGT\$LVTVSS (HC) DIQMTQSPSSLSASVGD\$RVTITCRASQDISNYLA\$WYQQKPGKVP KLLIFA\$ASTLQSGVPSRFGSGSGTDFTLTIS\$LPEDVATYYCQ NYNSGPP\$PFGPGTKVDIK (LC)	WO2016016278A3
mHTT	NI-302.33C11- PIMC	DFGMH (CDR-1 (HC)) LIWYDGGYKYYADSVKG (CDR-2 (HC)) HLEYCSRTTCYLGH (CDR-3 (HC)) RASQGISDYLA (CDR-1 (LC)) AASTLQT (CDR-2 (LC)) QQLKTYPYT (CDR-3 (LC)) QVQLVESGGGVVQPGNSLRL\$CAASGFRF\$DFGMHWVRQAPG KGLEWLALIWYDGGYKYYADSVKGRFTISRDN\$SKNTMFLQMN SLRAEDTAVYYCATHLEYCSRTTCYLGH\$WGQGT\$LVTVSS (HC) DIQLTQSP\$FLSASVGD\$TVTFTCRASQGISDYLA\$WFQQKPGIAPK LLIYA\$ASTLQTG\$VPSRFGSGSGTEFTLTIRSLQ\$SEDFGTYYCQQ LKTYPYTFGQGTKLEIK (LC)	WO2016016278A3

mHTT	NI-302.74C11-PIMC	GYFLH (CDR-1 (HC)) WINPNSGDTNYAE (CDR-2 (HC)) EAPDPGAETDV (CDR-3 (HC)) SGDAVPKQYIY (CDR-1 (LC)) KDTQRPS (CDR-2 (LC)) QSADSSATWV (CDR-3 (LC)) QVQLVQSGTEVQKPGASVKVSCKASGYSFTGYFLHWVRQAPG QGLEWMGWINPNSGDTNYAEKFRGRIMTRDTSVSTAHMELSS LRFDDTALYYCTREAPDPGAETDVWGQGTITVTVSS (HC) SYELTQPPSVSVSPGQTARITCSGDAVPKQYIYWYQQKPGQAPI LVIYKDTQRPSGIPERFSGSNSGTTVTLTITGVQADDEGDYYCQS ADSSATWVFGGGTKLTVL (LC)	WO2016016278A3
mHTT	NI-302.39G12-PIMC	NYAIT (CDR-1 (HC)) VIYRDGRYYGDSVRG (CDR-2 (HC)) AHGQYYYGVDV (CDR-3 (HC)) RSSQSLHNSNGYNYLD (CDR-1 (LC)) LSSNRPS (CDR-2 (LC)) MQSLQTFT (CDR-3 (LC)) EVQLVESGGGLVHPWGLRLSCAASGFSVSNYAITWVRRAPGK GLQYISVTYRDGRYYGDSVRGRFTISRDDSKNTLYLQMNSLRF EDTAVYYCARAHGQYYYGVVDVWGQGTITVTVSS (HC) DIVMTQSPLSLSVSPGEPASISCRSSQSLHNSNGYNYLDWYRQK PGQSPQLLIYLSSNRPSGVPDRFSASGSGTEFTLQISRVEAEDVG VYYCMQSLQTFTFGGGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.15E8	SYSMN (CDR-1 (HC)) YTSSRSNTKKYADSVKG (CDR-2 (HC)) AGDFGELLTGEgyyGMDV (CDR-3 (HC)) SGDELGDKYVG (CDR-1 (LC)) QDAKRPS (CDR-2 (LC)) QAWDSGTMV (CDR-3 (LC))	WO2016016278A3

mHTT	NI-302.64E5-PIMC	QAWMS (CDR-1 (HC)) RIKTKTEGEATDYAAPVRG (CDR-2 (HC)) TGVLAADVY (CDR-3 (HC)) KSSQSLFYSYNNENYLA (CDR-1 (LC)) WASTRES (CDR-2 (LC)) QYYSTPQT (CDR-3 (LC)) EVQLVESGGGLVKPGGSLRLSCAASGFTFDQAWMSWVRQVPG KGLEWVGRIKTKTEGEATDYAAPVRGRFTISRDDSEDTVFLQM NSLKTEDTALYYCTSTGVLAADVYWGQGLTVTVSS (HC) DIVMTQSPDSLAVSLGERATMTCKSSQSLFYSYNNENYLAWYQ QRPGQPPKLLIYWASTRESGVPRFSGSGSGTDFTLTISSLQAED VAVYYCQYYSTPQTFGQGTKVEIK (LC)	WO2016016278A3
mHTT	NI-302.72F10-PIMC	SYAMS (CDR-1 (HC)) DISGIGSNTYYADSVKG (CDR-2 (HC)) DRKRSGWYEQ (CDR-3 (HC)) RASQSISAYLG (CDR-1 (LC)) DASIRAT (CDR-2 (LC)) HQRSKWPLT (CDR-3 (LC)) EVQLVESGGGFVQPGGSLRLSCAASGFNFGSYAMSWVRQAPG KGLEWVSDISGIGSNTYYADSVKGRFTISRDNNDNTLYLDMSSL RAEDTARYYCAKDRKRSGWYEQWGQGLTVTVSS (HC) EIVLTQSPATLTSLSPGERATLSCRASQSISAYLGWYQKPGQAP RLLIYDASIRATGIPDRFSGSGSGTDFTLTISLPEPDSAVYYCHQ RSKWPLTFGGGTKVEIK (LC)	WO2016016278A3
IL-1 α	MABp1		The Lancet Oncology, 2017 15 (6): 656-66
IL-1 β	Canakinumab		Mabs, 2015, 7 (6): 1151-60
IL-1		[S/K/R]YDMS (CDR-1 (HC)) Y[I/V]S[S/H]GG[G/A]GTYYPD[T/S][V/A]KG (CDR-2 (HC)) GGV[T/Y]KG[Y/C]FD[V/E/L/M/Q/Y] (CDR-3 (HC)) RASGN[H/Y/W][N/G/T/Q/E/H/D/K][Y/W]L[T/A/N] (CDR-1 (LC)) [N/Q/D]K[T/N/I/E/S]L[A/M/E][D/E/S/A] (CDR-2 (LC)) Q[H/Q]FW[S/N/T/K/R/M][I/L]P[Y/A](T/I/N) (CDR-3 (LC))	WO2011143562A2

IL-RAP	CAN04	QVQLQDQSGPEELLKPGASVKISKAS GYAFSSSWMNWVKQRFGKGLEWIG RIYFGDGNTHYS GKFKGKATLTADK SSSIAYMQLSSSLTSEDSAVYFCQEG YLDPMQDYWGQGSTSVTVSS (HC) DIQMTQTTSLSASLGDRYTISCSA SQG!NNYLNWYQQKPDGTVKLLIH TSGLHAGVPSRFSSGSGTQVSLTI SNLEPEDVATYYCQQYSILPWTFG GGTKLEIKR (LC)	WO2015132602
IL-1 β	h1B12.4H4	DYGV(S CDR-1 (HC)) LIWGGGDTYNSPLKS (CDR-2 (HC)) QRTLWGYDLYGMDY (CDR-3 (HC)) ITSTDIDVDMN (CDR-1 (LC)) QGNTLRP (CDR-2 (LC)) LQSDNLPLT (CDR-3 (LC)) EVQLQESGGFLVPPSETLSLTCTVSGGSI DYGVSWIPQPPGKLENIQLINGGGDTYIN SPLKSPVTISVDTSKNQFSLKISSVTAADT AVYYCARQRTLWGYDLYGMDYWGQGT LVTVSS (HC) DIQMTQSPSSLSASVGRVTITCITSDID VDMNHWYQQKPGKAPKLLINQGNTLRPGVFS RFSGSGSGTDFTFTISSLQPEDFATYYCLQ SDNLPLTFGQGTKLEIK (LC)	WO2011047266A1
IL-1 α	H3D12	NYGMN (CDR-1 (HC)) WINTYTGESTYADDFKG (CDR-2 (HC)) GIYYYGSSYAMDY (CDR-3 (HC)) RASQDISNCLN (CDR-1 (LC)) YTSRLHS (CDR-2 (LC)) QQGKTLPPYA (CDR-3 (LC)) QIQLVQSGPEELKPGGETVKISKASGYTFR NYGMNWKQAPGKDLKRWINTYTGESTY ADDFKGRFAPSLETSASTAYLQINNLNED TATYFCARGIYYYGSSYAMDYWGQGSTVTV SS (HC) IQMTQTTSLSASLGDRVTISCRASQDIN CLAWYQQKPDGTVKLLIYYTSLRHSQVPER FECSGSGTQVSLTISNLEQEDIATYFCQQG KTLPPYAFGGGTKLEINR (LC)	WO 2010087972 A2

IL-1 β	Gevokizumab	TSGMGVG (CDR-1 (HC)) HIWWDGDESYNPSLK (CDR-2 (HC)) [A/V/F/K/N][R/K]YDPPWFVD (CDR-3 (HC)) RASQDISNYLS (CDR-1 (LC)) YTSKLHS (CDR-2 (LC)) LQGKMLPWT (CDR-3 (LC)) DIQMTQXTSSLSASXGDRVITIXCRASQDISNTLSWYQQKPKXXX VKLLIYYTSKLHSGVPSRFSGSGSGTDYXLTISNLEQEDIATYFC LQGKMLPWTFGGGKLEIK (HC) QVXLXESGPGXXKPSQTLSTCSFSGFSLSTSGMGVGWIRQPSG KGLEWLHAIWWDGDESYNPSLKXXLTISKDTSKNQVXLKITSV XXXDTAXYFCARXXYDPPWFVDWGQGLVTVSS (LC)	WO 2007002261 A2
IL-1 β	LY2189102		Diabetes Care 2013 Aug; 36(8): 2239- 2246.
IL-1 β	CYT013		
IL-1 β	sIL-1RII		
IL1RAP	Rilonacept		WO 2008051496 A2
IL-1RI	Anakinra		
IL-1RI	MEDI-8968		
IL-6	Tocilizumab		
IL-6	Sarilumab		
IL-6	Siltuximab		
IL-6	Vobarilizumab (ALX-0061)		Arthritis Research & Therapy 2015, 17:135
IL-6	Olokizumab		
IL-6	Elsilimomab		
IL-6	Clazakizumab		
IL-6	Sirukumab		

IL-6	Ab1	NYYVT (CDR-1 (HC)) IYGSDETAYATSAIG (CDR-2 (HC)) DDSSDWDAKFNL (CDR-3 (HC)) QASQSINNELS (CDR-1 (LC)) RASTLAS (CDR-2 (LC)) QQGYSLRNIDNA (CDR-3 (LC)) EVQLVESGGGLVQPGGSLRLSCAASGFSLSNYYVTWVRQAPGK GLEWVGIIYGSDETAYATSAIGRFTISRDN SKNTLYLQMNSLRA EDTAVYYCARDSSDWDAKFNLWGQGT LVT VSS (HC) AIQMTQSPSSLSASVGDRVTITCQASQSINNELSWYQQKPGKAP KLLIYRASTLASGVPSRFSGSGSGTDFTLTISSLQPDDFATYYCQ QGYSLRNIDNAFGGGTKVEIKR (LC)	WO 2011066371 A2; WO 2010065079 A2
TNF- α	Etanercept		
IL12 and or 23		DYY[M/L]H (CDR-1 (HC)) WIDPENGDT[E/Y/H/S]APKFQG (CDR-2 (HC)) [C/A/N/D/E/Q/G/H/I/L/P/V]KELRYFDV (CDR-3 (HC)) RA[S/P][Q/R]SISINLH (CDR-1 (LC)) FA[S/R]QS[I/T]S (CDR-2 (LC)) QQSNS[W/F]PLT (CDR-3 (LC)) TVAAPSVEIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVD NALQSGNSQESVTEQDSKDSSTYSLSSTLTLSKADYEKHKVYAC EVTHQGLSSPVTKSFNRGEC (HC) QPKAAPSVTLFPPSSEELQANKATLVCLISDFYPGAVTVAWKAD SSPVKAGVETTTPSKQSNKNKYAASSYLSTPEQWKSHRSYSCQ VTIEGSTVEKTVAPTECS (LC)	US 8563697 B2
IL12 and or 23	Ustekinumab		
IL12 and or 23	Briakinumab		

IL12 and IL23	R41F11	GFTFNNSWMT (CDR-1 (HC)) SITNTGGSTYYPDSVKG (CDR-2 (HC)) EDNSGYDY (CDR-3 (HC)) LASEDIYSNLA (CDR-1 (LC)) YANSLND (CDR-2 (LC)) QQNYYSPTT (CDR-3 (LC)) EVQLVESGGGLVQPGRSLKLSCLVASGFTFNNSWMTWIRQAPGK GLECVASITNTGGSTYYPDSVKGRFTISRDNASTLYLQMNSLR SEDTATYYCSREDNSGYDYWGQGVMVTVSS (HC) DIQMTQSPASLSASLGETVTIECLASEDIYSNLAWYQQKPGKSP QLLIYYANSLNDGVPSRFGSGSGTQYSLKINSLSQSEDSIYFCQ QNYYSPTTFGGGKLELKR (LC)	WO2008106134
IL12 and IL23	R8B10/HU8B1 0	GFDNFNSYMS (CDR-1 (HC)) DINSKSYNYATYYADSVKD (CDR-2 (HC)) HHSDYFEY (CDR-3 (HC)) LASEDIYNNLA (CDR-1 (LC)) HASSLQD (CDR-2 (LC)) LQDSEYPTT (CDR-3 (LC)) EVKLVESGGGLEQPKGSLKLSCTVSGFDNFNSYGMWVRQAPG KGLDLVADINSKSYNYATYYADSVKDRFTISRDDSQSMVYLEM DNLKTEDTALYYCTVHHSDYFEYWGQGVMVTVSS (HC) DIQMTQSPASLSASLGETVSIIECLASEDIYNNLAWYQQKPGKSP QLLIYHASSLQDGVPSRFGSGSGTQYSLKINSLESEDAATYFCL QDSEYPTTFGGGKLELKR (LC)	WO2008106134
IL12 and IL23	R3C11	GFSLSTYGMGVG (CDR-1 (HC)) NIWWDDDKYYNPSLKN (CDR-2 (HC)) IDAHPMGITTPDYVVDA (CDR-3 (HC)) KASQNVGSNVD (CDR-1 (LC)) KASNRYT (CDR-2 (LC)) MQSNSYPLT (CDR-3 (LC)) QVTLKESGPGILQPSQTLSTCTFSGFSLSTYGMGVGWIRQPSG KGLEWLNIWWDDDKYYNPSLKNRLTISKDTSNNQAFLKITNV DTADTATYYCARIDAHPMGITTPDYVVDAWGQGASVTVSS (HC) DIVMTQSPTSMISLGDRVTMNCKASQNVGSNVDWYQQKTGQ SPKLLIYKASNRYTGVPDRFTGSGSGTDFFTISNMQAEDLSVY YCMQSNSYPLTFGSGTKKEIKR (LC)	WO2008106134

IL12 and IL23	M20E5	GYTFTSYWIH (CDR-1 (HC)) EIDPSDSYTYYNQKFKD (CDR-2 (HC)) SLYDYDGVDPD (CDR-3 (HC)) HASQGIDNNIG (CDR-1 (LC)) HGTNLED (CDR-2 (LC)) VQYAQFPFT (CDR-3 (LC)) QVPLQQPGTELVKPGASVKLSCKASGYTFTSYWIHWVLQRPEQ GLEWIGEIDPSDSYTYYNQKFKDKATLTVDKSSTTAFLQLSSLT SEDSAVYYCARSLYDYDGVDPDWGQGTTLTVSA (HC) DILMTQSPSSMSVSLGDTVSTCHASQGIDNNIGWLQQKPGKSF KGLIYHGTNLEDGVPSRFGSGSGTDYSLTISSESEDFADYYCV QYAQFPFTFGGGTKLEIRR (LC)	WO2008106134
IL12 and IL23	Hu20D7	GYTFTNYAMN (CDR-1 (HC)) WINTYTGEPTYSDDFKG (CDR-2 (HC)) GGGYDEDYFDY (CDR-3 (HC)) KSSQSLFNSINQKTYLA (CDR-1 (LC)) FASTRES (CDR-2 (LC)) QQHYDTPWT (CDR-3 (LC)) QVQLVQSGAEVKKPGASVKVSCKASGYTFTNYAMNWVRQAP GQGLEWMGWINTYTGEPTYNEKFKGRVTMTDTSTSTAYMEL RSLRSDDTAVYYCARGGGYDEDYFDYWGQGTTLTVSS (HC) DIVMTQSPDSLAVSLGERATINCKSSQSLFNSINQKTYLAQYQQ KPGQPPKLLIYFASTRESGVPDRFSGSGSGTDFTLTISSLQAEDV AVYYCQQHYDTPWTFGQGTKVEIKRT (LC)	WO2008106134
Clq	ANX005 (ANX-M1)	GYHFTSYWMH (CDR-1 (HC)) VIHPNSGSINYNKES (CDR-2 (HC)) ERDSTEVLPMYD (CDR-3 (HC)) RASKSINKYLA (CDR-1 (LC)) SGSTLQS (CDR-2 (LC)) QQHNEYPLT (CDR-3 (LC)) QVQLQQPGAELVKPGASVKLSCKSSGYHFTSYWMHWVKQRP GQGLEWIGVIHPNSGSINYNKESKATLTVDKSSSTAYMQLSS LTSEDSAVYYCAGERDSTEVLPMYDYGQGTSTVTSS (HC) DVQITQSPSYLAASPGETTINCRASKSINKYLAWYQEKPGKTN KLLIYSGSTLQSGIPSRFSGSGSGTDFTLTISSELEPEDFAMYYCQQ HNEYPLTFGAGTKLELK (LC)	WO 2015006504 A1; EP 3019523 A4
Clq	JL-1		J Clin Invest. 2004; 114(5):679-688.

C5a	Eculizumab	GYIFSNIYWIQ (CDR-1 (HC)) EILPGSGSTEYTENFKD (CDR-2 (HC)) YFFGSSPNWYFDV (CDR-3 (HC)) GASENIYGALN (CDR-1 (LC)) GATNLAD (CDR-2 (LC)) QNVLNTPLT (CDR-3 (LC)) QVQLVQSGAEVKKPGASVKVSCKASGYIFSNIYWIQWVRQAPG QGLEWMGEILPGSGSTEYTENFKDRVTMTTRDTSTSTVYMELSS LRSEDTAVYYCARYFFGSSPNWYFDVWGQGLTVTVSS (HC) DIQMTQSPSSLSASVGDRTTITCGASENIYGALNWIYQQKPGKAP KLLIYGATNLADGVPSRFGSGSGTDFTLTISSLQPEDFATYYCQ NVLNTPLTFGQGTKVEIK (LC)	WO 2017044811 A1
APOE	15G8	FYAMS (CDR-1 (HC)) SLSRGGSTIYPDSVKG (CDR-2 (HC)) EGATALYAMDY (CDR-3 (HC)) RSSQSIVHSNGNTYLQ (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGSHPWT (CDR-3 (LC)) [E/Q]V[K/Q]L[V/Q]ESGGGLVKPGSLKLSAASGFTFSFYAMS WVRQTP[E/D]KRLEWVASLSRGGSTIYPDSVKGRTISR[D/S]N AKNTLYL[G/Q]MSSL[R/K]SEDTAMYYCAREGATALYAMDYW GQGTMTVTVSS (HC) DV[L/V]MTQSPLSLPVTLGQPASISCRSSQSIVHSNGNTYLQW[Y/ F]LQRPQSPR[L/R]LLYKVSNRFSQVDRFGSGSGTDFTLKISR VEAEDVGVYYCFQGSHPWTFGQGTKVEIK (LC)	WO2012109280 A2
APOE			US 20170218058 A1
APOE			WO 2013168174 A1
APOE			CA 2819679 A1

In a preferred embodiment, the multispecific binding molecule comprises at least a first binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or the V_L domains and/or V_H domains of B1B092, C2N 8E12, RG7345, 37D3-H9, 54C1-H11, 123E9, 24A11, Hu-37D3, Hu-125B11, Hu-94B2, DC8E8, Tau13, HT7, B1B076, 5A6 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%, even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains and a second binding site comprising a binding sequence selected from the group consisting of the combinations of CDR

sequences and/or V_L domains and/or V_H domains of BIIB092, C2N 8E12, RG7345, 37D3-H9, 54C1-H11, 123E9, 24A11, Hu-37D3, Hu-125B11, Hu-94B2, DC8E8, Tau13, HT7, BIIB076, 5A6 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%,
 5 even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains, wherein the first binding site and the second binding site are not identical.

In a further preferred embodiment, the multispecific binding molecule comprises at least a first
 10 binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or V_L domains and/or V_H domains of BIIB092, C2N 8E12, RG7345, 37D3-H9, 54C1-H11, 123E9, 24A11, Hu-37D3, Hu-125B11, Hu-94B2, DC8E8, Tau13, HT7, BIIB076, 5A6 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%,
 15 even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains and a second binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or V_L domains and/or V_H domains of PRX002/9E4, BIIB054, NI-202.3G12, NI-202.12F4, NI-202.3D8, GM285, GM37, BA1, BA2, BA3, BA4, 5C1, 9E4, Syn-211, H3C or fragments thereof
 20 having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%, even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains

Particularly preferred is a multispecific binding, wherein the first binding site is
 25 hu37D3-H9.v28 and the second binding site is M9E4, preferably wherein the multispecific binding molecule has a format of IgG-scFv, more preferably wherein hu37D3-H9.v28 is scFv.

In a preferred embodiment, the multispecific antibody comprises one binding site binding to human tau protein selected from the group comprising binding fragments of tau13 and binding
 30 fragments of 5A6 and one binding site binding to α-synuclein selected from the group comprising binding fragments of Syn211 and binding fragments of H3C.

In another preferred embodiment, the multispecific binding molecule comprises at least a first binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or V_L domains and/or V_H domains of BIIB092, C2N 8E12,
 35 RG7345, 37D3-H9, 54C1-H11, 123E9, 24A11, Hu-37D3, Hu-125B11, Hu-94B2, DC8E8, Tau13, HT7, BIIB076, 5A6 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%, even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains and a

second binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or V_L domains and/or V_H domains of ANX005 (ANX-M1) and JL-1 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%,
 5 even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains.

In another preferred embodiment, the multispecific antibody comprises one binding site binding to human tau protein selected from the group comprising binding fragments of tau13 and binding fragments of 5A6 and one binding site binding to C1q selected from the group comprising
 10 binding fragments of JL-1 and binding fragments produced by the hybridomas 23B6C8, 5B5C22, 12A5B7 and 4A4B11.

Also preferred is a multispecific binding molecule wherein the first binding site is Ab1 and the second binding site is ANX-M1, preferably wherein the multispecific binding molecule has a format of IgG-scFv.
 15

Also preferred is the multispecific antibody comprising one binding site binding to C1q selected from the group comprising binding fragments of JL-1 and binding fragments produced by the hybridomas 23B6C8, 5B5C22, 12A5B7 and 4A4B11 and one binding site binding to α -synuclein selected from the group comprising binding fragments of Syn211 and binding fragments of H3C.

It should be understood that for providing specificity it is preferred that a multispecific antibody according to the present invention would comprise the V_H or V_L of the specific antibodies as listed in the above table. Of course, alternatively an antibody may comprise the CDRs that are listed above for each and every binding compound listed, where it is preferred that a multispecific antibody comprises all 6 CDRs as presented above while the rest of the variable regions (and of course of the
 20 constant regions) may differ.
 25

The skilled person will also know that in the prior art a number of epitopes on the human tau protein have been identified that are suitable for targeting. The present invention includes multispecific binding molecules that specifically bind these epitopes in human (phosphorylated) tau protein.

30 Table 5: Epitopes of tau or phosphorylated (p)tau suitable for targeting.

Epitope	Reference
2-18	WO2014031694
9-18	WO2015081085
2-24	WO2016196726

7-9	WO2016137811
7-13	WO2014008404
15-24	WO2015081085
17-28	WO2015081085
19-33	WO2016196726
19-42	WO2016196726
24-46	CA2902026
25-30	WO2014008404/WO2015200806A2
28-44	WO2016196726
35-49	US20150252102A1
37-51	WO2016196726
P59+S61+E62+T63+D65+K67	WO2015197820
100-114	WO2016196726
109-123	WO2016196726
118-132	WO2016196726
150-158	WO2015081085
154-168	WO2016196726
172-177	WO2016196726
197-206	WO2017005732
217-231	WO2016196726
pT231	WO2012149334A2
268-273	WO2013041962A1/ WO2016079597A1
299-304	WO2013041962A1/WO2016079597A1
330-335	WO2013041962A1/ WO2016079597A1

362-367	WO2013041962A1/ WO2016079597A1
294-305	WO2013041962A1
306-320	WO2014008404
312-322	WO2016137811
D314+L315+K317	WO2015197820
L315+K317+P312	WO2015197820
337-343	US20150252102A1
387-397	US20130224287A1
pS396/pS404	WO2017009308A2
397-411	WO2016196726
pS413	WO2013180238A1
pS422	WO2015091656A1

Antibody Engineering

Further included are embodiments in which the antibodies of the invention and antigen-binding fragments thereof are engineered antibodies to include modifications to framework residues within the variable domains of the parental monoclonal antibody, *e.g.* to improve the properties of the antibody or fragment. Typically, such framework modifications are made to decrease the immunogenicity of the antibody or fragment. This is usually accomplished by replacing non-CDR residues in the variable domains (*i.e.* framework residues) in a parental (*e.g.* rodent) antibody or fragment with analogous residues from the immune repertoire of the species in which the antibody is to be used, *e.g.* human residues in the case of human therapeutics. Such an antibody or fragment is referred to as a "humanized" antibody or fragment. In some cases it is desirable to increase the affinity, or alter the specificity of an engineered (*e.g.* humanized) antibody. One approach is to "backmutate" one or more framework residues to the corresponding germline sequence. More specifically, an antibody or fragment that has undergone somatic mutation can contain framework residues that differ from the germline sequence from which the antibody is derived. Such residues can be identified by comparing the antibody or fragment framework sequences to the germline sequences from which the antibody or fragment is derived. Another approach is to revert to the original parental (*e.g.*, rodent) residue at one or more positions of the engineered (*e.g.* humanized) antibody, *e.g.* to restore binding affinity that may have been lost in the process of replacing the framework residues. (See, *e.g.*, U.S. Patent No. 5,693,762, U.S. Patent No. 5,585,089 and U.S. Patent No. 5,530,101.)

In certain embodiments, the antibodies of the invention and antigen-binding fragments thereof are engineered (e.g. humanized) to include modifications to in the framework and/or CDRs to improve their properties. Such engineered changes can be based on molecular modelling. A molecular model for the variable region for the parental (non-human) antibody sequence can be constructed to understand the structural features of the antibody and used to identify potential regions on the antibody that can interact with the antigen. Conventional CDRs are based on alignment of immunoglobulin sequences and identifying variable regions. Kabat et al., (1991) Sequences of Proteins of Immunological Interest, Kabat, et al.; National Institutes of Health, Bethesda, Md. ; 5th ed.; NIH Publ. No. 91-3242; Kabat Adv. Prot. Chem. 1978, 32:1-75; Kabat, et al., J. Biol. Chem. 1977, 252:6609-6616. Chothia and coworkers carefully examined conformations of the loops in crystal structures of antibodies and proposed hypervariable loops. Chothia, et al., Q J Mol. Biol. 1987, 196:901-917 or Chothia, et al., Nature 1989, 342:878-883. There are variations between regions classified as "CDRs" and "hypervariable loops". Later studies (Raghunathan et al, J. Mol Recog. 2012, 25, 3, 103-113) analyzed several antibody-antigen crystal complexes and observed that the antigen binding regions in antibodies do not necessarily conform strictly to the "CDR" residues or "hypervariable" loops. The molecular model for the variable region of the non-human antibody can be used to guide the selection of regions that can potentially bind to the antigen. In practice the potential antigen binding regions based on model differ from the conventional "CDR"s or "hyper variable" loops. Commercial scientific software such as Discovery Studio (BIOVIA, Dassault Systemes) can be used for molecular modeling. Human frameworks can be selected based on best matches with the non-human sequence both in the frameworks and in the CDRs. For FR4 (framework 4) in VH, VJ regions for the human germlines are compared with the corresponding non-human region. In the case of FR4 (framework 4) in VL, J-kappa and J-Lambda regions of human germline sequences are compared with the corresponding non-human region. Once suitable human frameworks are identified, the CDRs are grafted into the selected human frameworks. In some cases certain residues in the VL-VH interface can be retained as in the non-human (parental) sequence. Molecular models can also be used for identifying residues that can potentially alter the CDR conformations and hence binding to antigen. In some cases, these residues are retained as in the non-human (parental) sequence. Molecular models can also be used to identify solvent exposed amino acids that can result in unwanted effects such as glycosylation, deamidation and oxidation. Developability filters can be introduced early on in the design stage to eliminate/minimize these potential problems.

Another type of framework modification involves mutating one or more residues within the framework region, or even within one or more CDR regions, to remove T cell epitopes to thereby reduce the potential immunogenicity of the antibody. This approach is also referred to as "deimmunization" and is described in further detail in U.S. Patent No. 7,125,689.

In particular embodiments, it will be desirable to change certain amino acids containing exposed side-chains to another amino acid residue in order to provide for greater chemical stability of the final antibody, so as to avoid deamidation or isomerization. The deamidation of asparagine may occur on NG, DG, NG, NS, NA, NT, QG or QS sequences and result in the creation of an isoaspartic

acid residue that introduces a kink into the polypeptide chain and decreases its stability (isoaspartic acid effect). Isomerization can occur at DG, DS, DA or DT sequences. In certain embodiments, the antibodies of the present disclosure do not contain deamidation or asparagine isomerism sites.

For example, an asparagine (Asn) residue may be changed to Gln or Ala to reduce the potential for formation of isoaspartate at any Asn-Gly sequences, particularly within a CDR. A similar problem may occur at a Asp-Gly sequence. Reissner and Aswad *Cell. Mol. Life Sci.* 2003, 60:1281. Isoaspartate formation may debilitate or completely abrogate binding of an antibody to its target antigen. See, Presta J. *Allergy Clin. Immunol.* 2005, 116:731 at 734. In one embodiment, the asparagine is changed to glutamine (Gln). It may also be desirable to alter an amino acid adjacent to an asparagine (Asn) or glutamine (Gln) residue to reduce the likelihood of deamidation, which occurs at greater rates when small amino acids occur adjacent to asparagine or glutamine. See, Bischoff & Kolbe J. *Chromatog.* 1994, 662:261. In addition, any methionine residues (typically solvent exposed Met) in CDRs may be changed to Lys, Leu, Ala, or Phe or other amino acids in order to reduce the possibility that the methionine sulfur would oxidize, which could reduce antigen-binding affinity and also contribute to molecular heterogeneity in the final antibody preparation. *Id.* Additionally, in order to prevent or minimize potential scissile Asn-Pro peptide bonds, it may be desirable to alter any Asn-Pro combinations found in a CDR to Gln-Pro, Ala-Pro, or Asn-Ala. Antibodies with such substitutions are subsequently screened to ensure that the substitutions do not decrease the affinity or specificity of the antibody for its target, or other desired biological activity to unacceptable levels.

Table 6. Exemplary stabilizing CDR variants

CDR Residue	Stabilizing Variant Sequence
Asn-Gly (N-G)	Gln-Gly, Ala-Gly, or Asn-Ala (Q-G), (A-G), or (N-A)
Asp-Gly (D-G)	Glu-Gly, Ala-Gly or Asp-Ala (E-G), (A-G), or (D-A)
Met (typically solvent exposed) (M)	Lys, Leu, Ala, or Phe (K), (L), (A), or (F)
Asn (N)	Gln or Ala (Q) or (A)
Asn-Pro (N-P)	Gln-Pro, Ala-Pro, or Asn-Ala (Q-P), (A-P), or (N-A)

Antibody Engineering of the Fc region

The antibodies (*e.g.*, humanized antibodies) and antigen-binding fragments thereof disclosed herein can also be engineered to include modifications within the Fc region, typically to alter one or more properties of the antibody, such as serum, brain ISF or CSF half-life, complement
5 fixation, Fc receptor binding, and/or effector function (*e.g.*, antigen-dependent cellular cytotoxicity). Furthermore, the antibodies and antigen-binding fragments thereof disclosed herein can be chemically modified (*e.g.*, one or more chemical moieties can be attached to the antibody) or be modified to alter its glycosylation, again to alter one or more properties of the antibody or fragment. Each of these embodiments is described in further detail below. The numbering of residues in the Fc
10 region is that of the EU index of Kabat.

The antibodies and antigen-binding fragments thereof disclosed herein also include antibodies and fragments with modified (or blocked) Fc regions to provide altered effector functions. See, *e.g.*, U.S. Pat. No. 5,624,821; WO2003/086310; WO2005/120571; WO2006/0057702. Such
15 modifications can be used to enhance or suppress various reactions of the immune system, with possible beneficial effects in diagnosis and therapy. Alterations of the Fc region include amino acid changes (substitutions, deletions and insertions), glycosylation or deglycosylation, and adding multiple Fc regions. Changes to the Fc can also alter the half-life of antibodies in therapeutic
antibodies, enabling less frequent dosing and thus increased convenience and decreased use of material. See Presta J. Allergy Clin. Immunol. 2005, 116:731 at 734-35.

20 In one embodiment, the Fc region of the antibody or antigen-binding fragment of the invention is modified to prevent possible interactions with the neonatal Fc receptor. Such modifications can be used to decrease efflux of the antibody or antigen-binding fragment out of the brain and thus increase brain half-life. (Cooper, P.R., Brain Research, 2013, 1534, 13).

In one embodiment, the antibody or antigen-binding fragment of the invention is an
25 IgG4 isotype antibody or fragment comprising a Serine to Proline mutation at a position corresponding to position 228 (S228P; EU index) in the hinge region of the heavy chain constant region. This mutation has been reported to abolish the heterogeneity of inter-heavy chain disulfide bridges in the hinge region (Angal S. et al., Mol Immunol. 1993, 30: 105-108; position 241 is based on the Kabat numbering system).

30 In one embodiment of the invention, the hinge region of CH₁ is modified such that the number of cysteine residues in the hinge region is increased or decreased. This approach is described further in U.S. Patent No. 5,677,425. The number of cysteine residues in the hinge region of CH₁ is altered, for example, to facilitate assembly of the light and heavy chains or to increase or decrease the stability of the antibody.

35 In another embodiment, the Fc hinge region of an antibody or antigen-binding fragment of the invention is mutated to decrease the biological half-life of the antibody or fragment. More

specifically, one or more amino acid mutations are introduced into the CH₂-CH₃ domain interface region of the Fc-hinge fragment such that the antibody or fragment has impaired Staphylococcal protein A (SpA) binding relative to native Fc-hinge domain SpA binding. This approach is described in further detail in U.S. Patent No. 6,165,745.

5 In another embodiment, the antibody or antigen-binding fragment of the invention is modified to increase its biological half-life. Various approaches are possible. For example, one or more of the following mutations can be introduced: T252L, T254S, T256F, as described in U.S. Patent No. 6,277,375. Alternatively, to increase the biological half-life, the antibody can be altered within the CH₁ or C_L region to contain a salvage receptor binding epitope taken from two loops of a CH₂ domain
10 of an Fc region of an IgG, as described in U.S. Patent Nos. 5,869,046 and 6,121,022.

 In yet other embodiments, the Fc region is altered by replacing at least one amino acid residue with a different amino acid residue to alter the effector function(s) of the antibody or antigen-binding fragment. For example, one or more amino acids selected from amino acid residues 233, 234, 235, 236, 237, 268, 269, 270, 254, 254, 294, 297, 298, 300, 318, 320, 322, 327, 329, 331 can be replaced
15 with a different amino acid residue such that the antibody has an altered affinity for an effector ligand and retains the antigen-binding ability of the parent antibody. The effector ligand to which affinity is altered can be, for example, an Fc receptor or the C₁ component of complement. This approach is described in further detail in U.S. Patent Nos. 5,624,821 and 5,648,260. Ugurlar, D., Science, 2018, 359, 6377, 794.

20 In another example, one or more amino acids selected from amino acid residues 329, 331 and 322 can be replaced with a different amino acid residue such that the antibody has altered C1q binding and/or reduced or abolished complement dependent cytotoxicity (CDC). This approach is described in further detail in U.S. Patent No. 6,194,551.

 In another example, one or more amino acid residues within amino acid positions 231
25 and 239 are altered to thereby alter the ability of the antibody to fix complement. This approach is described further in PCT Publication WO 94/29351.

 In yet another example, the Fc region is modified to decrease the ability of the antibody or antigen-binding fragment of the invention to mediate antibody dependent cellular cytotoxicity (ADCC) and/or to decrease the affinity of the antibody or fragment for an Fcγ receptor by modifying
30 one or more amino acids at the following positions: 238, 239, 243, 248, 249, 252, 254, 255, 256, 258, 264, 265, 267, 268, 269, 270, 272, 276, 278, 280, 283, 285, 286, 289, 290, 292, 293, 294, 295, 296, 298, 301, 303, 305, 307, 309, 312, 315, 320, 322, 324, 326, 327, 329, 330, 331, 333, 334, 335, 337, 338, 340, 360, 373, 376, 378, 382, 388, 389, 398, 414, 416, 419, 430, 434, 435, 437, 438 or 439. This approach is described further in PCT Publication WO 00/42072. Moreover, the binding sites on human IgG1 for
35 FcγR1, FcγRII, FcγRIII and FcRn have been mapped and variants with improved binding have been described (*see* Shields et al. J. Biol. Chem. 2001, 276:6591-6604).

In one embodiment of the invention, the Fc region is modified to decrease the ability of the antibody of the invention to mediate effector function and/or to increase anti-inflammatory properties by modifying residues 243 and 264. In one embodiment, the Fc region of the antibody or fragment is modified by changing the residues at positions 243 and 264 to alanine. In one
5 embodiment, the Fc region is modified to decrease the ability of the antibody or fragment to mediate effector function and/or to increase anti-inflammatory properties by modifying residues 243, 264, 267 and 328.

Antibody Physical Properties

The antibodies and antigen-binding fragments thereof disclosed herein may further
10 contain one or more glycosylation sites in either the light or heavy chain immunoglobulin variable region. Such glycosylation sites may result in increased immunogenicity of the antibody or fragment or an alteration of the pK of the antibody due to altered antigen-binding (Marshall et al. Annu Rev Biochem 1972, 41:673-702; Gala and Morrison, J. Immunol 2004, 172:5489-94; Wallick et al. J Exp Med 1988, 168:1099-109; Spiro Glycobiology, 2002, 12:43R-56R; Parekh et al. Nature 1985, 316:452-7;
15 Mimura et al., Mol Immunol 2000, 37:697-706). Glycosylation has been known to occur at motifs containing an N-X-S/T sequence.

Each antibody or antigen-binding fragment will have a unique isoelectric point (pI), which generally falls in the pH range between 6 and 9.5. The pI for an IgG1 antibody typically falls within the pH range of 7-9.5 and the pI for an IgG4 antibody typically falls within the pH range of 6-
20 8.

Each antibody or antigen-binding fragment will have a characteristic melting temperature, with a higher melting temperature indicating greater overall stability *in vivo* (Krishnamurthy, R and Manning, M.C. Curr Pharm Biotechnol 2002, 3:361-71). In general, the T_{M1} (the temperature of initial unfolding) may be greater than 60°C, greater than 65°C, or greater than
25 70°C. The melting point of an antibody or fragment can be measured using differential scanning calorimetry (Chen et al., Pharm Res 2003, 20:1952-60; Ghirlando et al. Immunol Lett, 1999, 68:47-52) or circular dichroism (Murray et al., J. Chromatogr Sci 2002, 40:343-9).

In a further embodiment, antibodies and antigen-binding fragments thereof are selected that do not degrade rapidly. Degradation of an antibody or fragment can be measured using capillary
30 electrophoresis (CE) and MALDI-MS (Alexander, A.J. and Hughes, D.E. Anal Chem, 1995, 67:3626-32).

In a further embodiment, antibodies and antigen-binding fragments thereof are selected that have minimal aggregation effects, which can lead to the triggering of an unwanted immune response and/or altered or unfavorable pharmacokinetic properties. Generally, antibodies and
35 fragments are acceptable with aggregation of 25% or less, 20% or less, 15% or less, 10% or less, or 5%

or less. Aggregation can be measured by several techniques, including size-exclusion column (SEC), high performance liquid chromatography (HPLC), and light scattering.

Antibody Conjugates

5 The antibodies and antigen-binding fragments thereof disclosed herein may also be conjugated to a chemical moiety. The chemical moiety may be, *inter alia*, a polymer or a compound that enables transport over the blood brain barrier (BBB). In particular embodiments, the chemical moiety is a polymer which increases the half-life of the antibody or fragment in the body of a subject. Suitable polymers include, but are not limited to, hydrophilic polymers which include but are not limited to polyethylene glycol (PEG) (*e.g.*, PEG with a molecular weight of 2 kDa, 5 kDa, 10 kDa, 12
10 kDa, 20 kDa, 30 kDa or 40 kDa), dextran and monomethoxypolyethylene glycol (mPEG). Antibodies or antigen-binding fragments thereof may for example be PEGylated to increase its biological (*e.g.* serum) half-life. To PEGylate an antibody or fragment, the antibody or fragment, typically is reacted with a reactive form of polyethylene glycol (PEG), such as a reactive ester or aldehyde derivative of PEG, under conditions in which one or more PEG groups become attached to the antibody or antibody
15 fragment. In particular embodiments, the PEGylation is carried out via an acylation reaction or an alkylation reaction with a reactive PEG molecule (or an analogous reactive water-soluble polymer). As used herein, the term "polyethylene glycol" is intended to encompass any of the forms of PEG that have been used to derivatize other proteins, such as mono (C1-C10) alkoxy- or aryloxy-polyethylene glycol or polyethylene glycol-maleimide. In certain embodiments, the antibody or fragment to be
20 PEGylated is an aglycosylated antibody or fragment. Methods for PEGylating proteins are known in the art and can be applied to the antibodies of the invention. *See, e.g.*, EP 0 154 316 and EP 0 401 384. Lee, et al., *Bioconj. Chem.* 1999, 10:973-981 discloses PEG conjugated single-chain antibodies. Wen, et al., *Bioconj. Chem.* 2001, 12:545-553 disclose conjugating antibodies with PEG which is attached to a radiometal chelator (diethylenetriaminopentaacetic acid (DTPA)).

25 The antibodies and antigen-binding fragments thereof, especially when used for diagnostic purposes, disclosed herein may also be conjugated with labels such as ^{99}Tc , ^{90}Y , ^{111}In , ^{32}P , ^{14}C , ^{125}I , ^3H , ^{131}I , ^{11}C , ^{15}O , ^{13}N , ^{18}F , ^{35}S , ^{51}Cr , ^{57}To , ^{226}Ra , ^{60}Co , ^{59}Fe , ^{57}Se , ^{152}Eu , ^{67}Cu , ^{217}Cl , ^{211}At , ^{212}Pb , ^{47}Sc , ^{109}Pd , ^{234}Th , and ^{40}K , ^{157}Gd , ^{55}Mn , ^{52}Tr , and ^{56}Fe .

30 The antibodies and antigen-binding fragments disclosed herein, especially when used for diagnostic purposes, may also be conjugated with fluorescent or chemiluminescent labels, including fluorophores such as rare earth chelates, fluorescein and its derivatives, rhodamine and its derivatives, isothiocyanate, phycoerythrin, phycocyanin, allophycocyanin, o-phthalaldehyde, fluorescamine, ^{152}Eu , dansyl, umbelliferone, luciferin, luminal label, isoluminal label, an aromatic acridinium ester label, an imidazole label, an acridinium salt label, an oxalate ester label, an
35 aequorin label, 2,3-dihydrophthalazinediones, biotin/avidin, spin labels and stable free radicals.

Any method known in the art for conjugating the antibodies and antigen-binding fragments thereof of the invention to the various moieties may be employed, including those methods described by Hunter, et al., *Nature* 1962, 144:945; David, et al., *Biochemistry* 1974, 13:1014; Pain, et al., *J. Immunol. Meth.* 1981, 40:219; and Nygren, J., *Histochem. and Cytochem.* 1982, 30:407.

5 Methods for conjugating antibodies and fragments are conventional and very well known in the art.

Antibodies or other polypeptides may be immobilized onto a variety of solid supports for use in assays. Solid phases that may be used to immobilize specific binding members include those developed and/or used as solid phases in solid phase binding assays. Examples of suitable solid phases include membrane filters, cellulose-based papers, beads (including polymeric, latex and
10 paramagnetic particles), glass, silicon wafers, microparticles, nanoparticles, TentaGels, AgroGels, PEGA gels, SPOCC gels, and multiple-well plates. An assay strip could be prepared by coating the antibody or a plurality of antibodies in an array on solid support. This strip could then be dipped into the test sample and then processed quickly through washes and detection steps to generate a measurable signal, such as a colored spot. Antibodies or other polypeptides may be bound to specific
15 zones of assay devices either by conjugating directly to an assay device surface, or by indirect binding. In an example of the later case, antibodies or other polypeptides may be immobilized on particles or other solid supports, and that solid support immobilized to the device surface.

Biological assays require methods for detection, and one of the most common methods for quantitation of results is to conjugate a detectable label to a protein or nucleic acid that has affinity
20 for one of the components in the biological system being studied. Detectable labels may include molecules that are themselves detectable (e.g., fluorescent moieties, electrochemical labels, metal chelates, etc.) as well as molecules that may be indirectly detected by production of a detectable reaction product (e.g., enzymes such as horseradish peroxidase, alkaline phosphatase, etc.) or by a specific binding molecule which itself may be detectable (e.g., biotin, digoxigenin, maltose,
25 oligohistidine, 2,4-dinitrobenzene, phenylarsenate, ssDNA, dsDNA, etc.).

Preparation of solid phases and detectable label conjugates often comprise the use of chemical cross-linkers. Cross-linking reagents contain at least two reactive groups, and are divided generally into homofunctional cross-linkers (containing identical reactive groups) and heterofunctional cross-linkers (containing non-identical reactive groups). Homobifunctional cross-
30 linkers that couple through amines, sulfhydryls or react non-specifically are available from many commercial sources. Maleimides, alkyl and aryl halides, α -haloacyls and pyridyl disulfides are thiol reactive groups. Maleimides, alkyl and aryl halides, and α -haloacyls react with sulfhydryls to form thiol ether bonds, while pyridyl disulfides react with sulfhydryls to produce mixed disulfides. The pyridyl disulfide product is cleavable. Imidoesters are also very useful for protein-protein cross-links.
35 A variety of heterobifunctional cross-linkers, each combining different attributes for successful conjugation, are commercially available.

Intracellular delivery of binding molecules

In some cases, the multispecific binding molecule targets a pathological target that is located inside of the cell. Hence, intracellular delivery of the multispecific binding molecule is required. Multispecific binding molecules such as multispecific antibodies often lack the ability to efficiently penetrate cellular membranes due to their large size and hydrophilicity. Various formats are published in literature to improve delivery of sufficient amounts of intact binding molecules into cells.

For example, cytotransmabs are antibodies comprising a cytosol penetrating light chain variable domain paired with a heavy chain variable domain of a therapeutic antibody. Internalization of the cytotransmabs was achieved through interaction with heparin sulfate proteoglycan (HSPS), resulting in clathrin-mediated endocytic pathway. The internalized cytotransmabs did not show noticeable cytotoxicity, which indicates that initialization takes place via a physical endocytosis pathway without disrupting the membrane. Hence, cytotransmabs provide a suitable method for improving internalization of a therapeutic antibody. (Choi, D-K, mAbs, 2014, 6:6, 1402). Alternatively, a fusion protein of the IgG1-Fc domain with Cre recombinase also allowed permeation of the cell membrane. (Marshall, A.J., mAbs, 2014, 6:4, 953)

Furthermore, intracellular delivery vehicles that are able to transport antibodies into the cell have been reported. A fusion protein consisting of Protein A domain B and the self-assembling Hex domain forms a nanocarrier that tightly binds antibodies. This vehicle-antibody complex shows improved internalization compared to soluble antibodies and indeed enhances the delivery of the antibodies to the cytosol. (In Lim, S., J. Control. Release, 2017).

Another possibility is to use a therapeutic delivery vehicle comprising a replication defective virus, such as an adeno-associated virus (AAV) vector. These vectors can carry genes, for example genes encoding multispecific binding molecules or fragments thereof and enable administration of those genes into the cell. Therefore, these vehicles are particularly useful for gene-therapy. The use of AAV as delivery vehicles has several advantages. For example, AAV is a virus that infects cells but is not known to cause a disease and does not exhibit significant immunogenicity. These therapeutic delivery vehicles are also suitable for enabling expression of multispecific binding molecules of the invention directly into the targeted cell. Hence, a specific cell targeted therapy effected by providing a subject a therapeutic delivery vehicle according to the invention may be used in therapies against NDDs.

Whereas AAV packages a single strand of DNA and must wait for its second strand to be synthesized, scAAV packages two shorter strands that are complementary to each other. By avoiding second-strand synthesis, scAAV can express more quickly, although as a caveat, scAAV can only encode half of the already limited capacity of AAV. This methodology is especially suitable for expressing self-complimentary antibody fragments of the invention.

Blood brain barrier (BBB) permeation

One of the major hurdles in the development of effective treatments for disorders affecting the central nervous system, such as NDDs, is the low efficiency to deliver the therapeutics into the brain. For example, in antibody-based treatment only 0.1% of circulating antibodies end up in the brain. The main reason for this low efficiency is the presence of the BBB which tightly controls the transfer of substances from the blood to the brain tissue and *vice versa*. It consists of tight junctions enclosing the brain endothelial and epithelial cells, which hinders the paracellular passage of molecules.

Molecules that are allowed to passage the BBB can be utilized to transport cargo across this barrier. Such transport can be achieved either specifically, by targeting transporters that are highly expressed on the BBB, or aspecifically. The family of cell penetrating peptides (CPPs), comprised of short amphiphatic or cationic sequences, constitute a group of peptides that enable aspecific passage of cargo through the BBB.

Alternatively, BBB shuttles have been developed that are able to pass the BBB through the targeting of receptors present on the BBB (receptor-mediated transport). Ideally, such receptors should be highly expressed on the luminal side of brain vasculature with respect to other tissues, have a high capacity for transcytosis, broad substrate scope and high turnover. In addition, the physiological role of the transporter should remain unaffected. Examples of such receptors include but are not limited to transferrin receptor (TfR), insulin receptor (IR), leptin receptor, low density lipoproteins (LDLRs), lipoprotein receptor-related protein (LRP), CD98hc, n4ChR, KCa channel, GSH, GM1, AMT, FC5, IGF1 receptor, IGF2 receptor, FCGRT, Scavenger receptor class B, basigin, LRP, the melanocortin receptor, the nicotinic acetylcholine receptor, the VACM-1 receptor, IGFR, EPCR, EGFR, TNFR, M6PR, the lipoprotein receptor, NCAM, LIFR, MRP1, AchR, DTr, the glutathione transporter, SR-B1, MYOF, TFRC, ECE1, PVR, CDC50A, SCARF1, MRC1, HLA-DRA, RAMP2, VLDLR, STAB1, TLR9, CXCL16, NTRK1, CD74, DPP4, endothelial growth factor receptors 1, 2 and 3, the glucocorticoid receptor, the ionotropic glutamate receptor, the M3 receptor, the aryl hydrocarbon receptor, GLUT-1, inositol-1, 4,5-trisphosphate (IP3) receptor, the N-methyl- D-aspartate receptor, S1 P1, the P2Y receptor, TMEM30A, and RAGE. These receptors can be targeted with either peptides or proteins, such as antibodies/antibody-fragments.

Peptides targeting these receptors were found exploiting phage display, or can be derived from natural proteins such as apolipoproteins or hormones, or from exogenous proteins like neurotoxins or viruses. Examples of peptide-based BBB shuttles include but are not limited to RDP, KC2S, , Angiopep-2, ApoB(3371-3409), ApoE(159-167), peptide-22, THR, THR *retro enantio*, CRT, Leptin30, RVG29, CDX, Apamin, MiniAP-4, GSH, G23, g7, TGN, TAT(47-57), SynB1, (phenylproline)4-NH2, Diketopiperazine (Table 8). These shuttle components can be easily attached to the multispecific binding molecules of the present invention by using simple chemical cross-linking reagents as described above. For a full description of peptide-based BBB shuttles, including sequences of the

shuttles, active transporters, passive diffusion and the *retro enantio* approach see Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707.

One challenge associated with the process of intracellular delivery of macromolecules using CPP's is the efficient endosomal release of the macromolecule into the cytoplasm. It was found that linking the macromolecule/CPP complex to a hydrophobic motif led to significant enhancement of release of the complex from the endosomes. This enhanced release is due to incorporation of the hydrophobic motif into the lipid bilayer of the endosome, leading to destabilization of the membrane and consequent release of the macromolecule into the cytoplasm. (Lönn, P., Scientific reports, 2016, 6:32301). Alternatively, proteins such as antibodies or antibody-fragments can be used to enhance passage of the BBB. Increased permeation of the BBB has been demonstrated for a number of bispecific antibodies comprising one binding site targeting the TfR, insulin receptor or glycosylated protein (Cdc50A) and these antibodies are currently being investigated in preclinical trials. (Neves, V., Trends in biotechnology). Different antibodies or antibody-fragments were found to be suitable to enhance influx from the blood to the brain tissue. These examples (Table 8) are therefore also of interest in the current invention.

When binding molecules do not bind receptors directly but through the binding of a carbohydrate-ligand, absorptive-mediated transcytosis (AMT) can take place. This form of transcytosis was exemplified by the antibody FC5 that binds α 2,3-linked sialic acid groups on Cdc50A. Binding of FC5 to these groups resulted into enhanced influx of FC5 into the brain.

It was additionally demonstrated that different antibody formats allow different mechanisms of uptake in the brain. For example, antibodies with high affinity for the target receptor are less likely to be released afterwards. Careful selection of the antibody format, additionally enables the adjustment of certain physical properties, such as toxicity. Toxic side effects of an anti TfR-Ab could be significantly reduced when the equivalent antibody, containing only one Fab arm, was administered. No detectable cytotoxicity was observed when the anti TfR Ab was linked to the C-terminal of the anti-Ab antibody, primarily because binding to transferrin receptors in the periphery was prevented due to sterical hindrance. Moreover, it was found that when two scFv of a TfR antibody were recombinantly fused to the C-terminal of the light chain of an A β antibody, an antibody was obtained showing increased uptake into the brain. The design, comprising a short linker connecting the scFv's to the A β antibody, enabled monovalent binding to the TfR. Multivalent binding was prevented due to sterical hindrance. As a consequence, the antibody was released more easily, although the likelihood of TfR-binding and thus brain uptake, was doubled compared to antibodies comprising a monovalent binding site for TfR. (Weber, F., Cell Reports, 2017, 22, 149–162; Hultqvist, G., Theranostics 2017, 7(2), 308).

For reviews on strategies crossing the BBB using antibodies or antibody-fragments see Neves, V, Trends in biotechnology, Partridge, W., Biodrugs, 2017. All the strategies that are discussed in this reference can also be used for the present invention.

Besides improving the uptake of the multispecific binding molecules in the brain, extending the brain half life of the multispecific binding molecule is equally important. One way to achieve this goal would be to increase the influx of the multispecific binding molecules into the brain. Such an effect can be achieved by using peptide- or protein-based shuttles to enable passage of the BBB as outlined above. Alternatively, smaller multispecific binding molecules such as multispecific Fab fragments, might be more successful in passing the BBB transcellularly. (Finke, J.M., Human Antibodies, 2016).

Another way to improve the influx of multispecific binding molecules in the brain, is by giving a patient ultrasonic pulses that temporarily disrupt part of the BBB via microbubble cavitation and allows passage of substances, such as drugs or endogenous proteins. To minimize side-effects, the timing and location of the ultrasonic pulse is important. Enhanced brain and neuronal uptake of a single-chain variable fragment of an anti-tau antibody was achieved, leading to decreased tau phosphorylation in a tau transgenic mouse model.

However, apart from improving influx, reducing the efflux of the multispecific binding molecules could also contribute to an extended half life of the binding molecule. Reducing efflux from the brain can be achieved by modifying the Fc region of the antibody or antibody fragment. Moreover, it was demonstrated that sialylation of glycans expressed on the Fab fragment can also extend the serum half life of the antibody or antibody fragment. (Finke, J.M., Human Antibodies, 2016).

In a preferred embodiment of the present invention, the multispecific binding molecule additionally comprises an additional binding specificity binding a further binding target selected from the group comprising transferrin receptor (TfR), insulin receptor (IR), leptin receptor, low density lipoproteins (LDLRs), lipoprotein receptor-related protein (LRP), CD98hc, n4ChR, KCa channel, GSH, GM1, AMT, IGF1 receptor, IGF2 receptor, FCGRT, Scavenger receptor class B, basigin, LRP, the melanocortin receptor, the nicotinic acetylcholine receptor, the VACM-1 receptor, IGFR, EPCR, EGFR, TNFR, M6PR, the lipoprotein receptor, NCAM, LIFR, MRP1, AchR, DTr, the glutathione transporter, SR-B1, MYOF, TFRC, ECE1, PVR, CDC50A, SCARF1, MRC1, HLA-DRA, RAMP2, VLDLR, STAB1, TLR9, CXCL16, NTRK1, CD74, DPP4, endothelial growth factor receptors 1, 2 and 3, the glucocorticoid receptor, the ionotropic glutamate receptor, the M3 receptor, the aryl hydrocarbon receptor, GLUT-1, inositol-1, 4,5-trisphosphate (IP3) receptor, the N-methyl- D-aspartate receptor, S1 P1, the P2Y receptor, TMEM30A, and RAGE to enable passage of the multispecific binding molecule through the BBB.

Preferably, the additional binding specificity is an antibody or antibody-fragment. In one aspect the binding site specifically targets a BBB receptor selected from the group comprising transferrin receptor, insulin receptor, CD98hc, leptin and low density lipoproteins.

All combinations are possible and accordingly the present invention comprises a plurality of different multispecific binding molecules, including but not limited to the combinations listed below, where the codes A1-A5, B1-B16 and C1-C6 are described in the Tables 1-2 & 7.

Table 7: Transporters used to passage the BBB

Entry	Target
C1	transferrin receptor
C2	insulin receptor
C3	leptin receptor
C4	low density lipoproteins
C5	CD98hc
C6	ApoE(159-167)

5	A1+A2+C1, A1+A3+C1, A1+A4+C1, A1+A5+C1, A2+A3+C1, A2+A4+C1, A3+A4+C1, A4+A5+C1,
	A1+B1+C1, A1+B2+C1, A1+B3+C1, A1+B4+C1, A1+B5+C1, A1+B6+C1, A1+B7+C1, A1+B8+C1,
	A2+B1+C1, A2+B2+C1, A2+B3+C1, A2+B4+C1, A2+B5+C1, A2+B6+C1, A2+B7+C1, A2+B8+C1,
	A3+B1+C1, A3+B2+C1, A3+B3+C1, A3+B4+C1, A3+B5+C1, A3+B6+C1, A3+B7+C1, A3+B8+C1,
	A4+B1+C1, A4+B2+C1, A4+B3+C1, A4+B4+C1, A4+B5+C1, A4+B6+C1, A4+B7+C1, A4+B8+C1,
10	A5+B1+C1, A5+B2+C1, A5+B3+C1, A5+B4+C1, A5+B5+C1, A5+B6+C1, A5+B7+C1, A5+B8+C1,
	A1+A2+C2, A1+A3+C2, A1+A4+C2, A1+A5+C2, A2+A3+C2, A2+A4+C2, A3+A4+C2, A4+A5+C2,
	A1+B1+C2, A1+B2+C2, A1+B3+C2, A1+B4+C2, A1+B5+C2, A1+B6+C2, A1+B7+C2, A1+B8+C2,
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- A4+B1+C5, A4+B2+C5, A4+B3+C5, A4+B4+C5, A4+B5+C5, A4+B6+C5, A4+B7+C5, A4+B8+C5,
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- $A2+A4+B7+C1$, $A2+A4+B8+C1$, $A3+A4+B1+C1$, $A3+A4+B2+C1$, $A3+A4+B3+C1$, $A3+A4+B4+C1$,
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	A1+A2+B1+C3, A1+A2+B2+C3, A1+A2+B3+C3, A1+A2+B4+C3, A1+A2+B5+C3, A1+A2+B6+C3, A1+A2+B7+C3, A1+A2+B8+C3, A1+A3+B1+C3, A1+A3+B2+C3, A1+A3+B3+C3, A1+A3+B4+C3, A1+A3+B5+C3, A1+A3+B6+C3, A1+A3+B7+C3, A1+A3+B8+C3, A1+A4+B1+C3, A1+A4+B2+C3, A1+A4+B3+C3, A1+A4+B4+C3, A1+A4+B5+C3, A1+A4+B6+C3, A1+A4+B7+C3, A1+A4+B8+C3,
5	A1+A5+B1+C3, A1+A5+B2+C3, A1+A5+B3+C3, A1+A5+B4+C3, A1+A5+B5+C3, A1+A5+B6+C3, A1+A5+B7+C3, A1+A5+B8+C3, A1+B1+B2+C3, A1+B1+B3+C3, A1+B1+B4+C3, A1+B1+B5+C3, A1+B1+B6+C3, A1+B1+B7+C3, A1+B1+B8+C3, A1+B2+B3+C3, A1+B2+B4+C3, A1+B2+B5+C3, A1+B2+B6+C3, A1+B2+B7+C3, A1+B2+B8+C3, A1+B3+B4+C3, A1+B3+B5+C3, A1+B3+B6+C3, A1+B3+B7+C3, A1+B3+B8+C3, A1+B4+B5+C3, A1+B4+B6+C3, A1+B4+B7+C3, A1+B4+B8+C3,
10	A1+B5+B6+C3, A1+B5+B7+C3, A1+B5+B8+C3, A1+B6+B7+C3, A1+B6+B8+C3, A1+B7+B8+C3, A2+B1+B2+C3, A2+B1+B3+C3, A2+B1+B4+C3, A2+B1+B5+C3, A2+B1+B6+C3, A2+B1+B7+C3, A2+B1+B8+C3, A2+B2+B3+C3, A2+B2+B4+C3, A2+B2+B5+C3, A2+B2+B6+C3, A2+B2+B7+C3, A2+B2+B8+C3, A2+B3+B4+C3, A2+B3+B5+C3, A2+B3+B6+C3, A2+B3+B7+C3, A2+B3+B8+C3, A2+B4+B5+C3, A2+B4+B6+C3, A2+B4+B7+C3, A2+B4+B8+C3, A2+B5+B6+C3, A2+B5+B7+C3,
15	A2+B5+B8+C3, A2+B6+B7+C3, A2+B6+B8+C3, A2+B7+B8+C3, A3+B1+B2+C3, A3+B1+B3+C3, A3+B1+B4+C3, A3+B1+B5+C3, A3+B1+B6+C3, A3+B1+B7+C3, A3+B1+B8+C3, A3+B2+B3+C3, A3+B2+B4+C3, A3+B2+B5+C3, A3+B2+B6+C3, A3+B2+B7+C3, A3+B2+B8+C3, A3+B3+B4+C3, A3+B3+B5+C3, A3+B3+B6+C3, A3+B3+B7+C3, A3+B3+B8+C3, A3+B4+B5+C3, A3+B4+B6+C3, A3+B4+B7+C3, A3+B4+B8+C3, A3+B5+B6+C3, A3+B5+B7+C3, A3+B5+B8+C3, A3+B6+B7+C3,
20	A3+B6+B8+C3, A3+B7+B8+C3, A4+B1+B2+C3, A4+B1+B3+C3, A4+B1+B4+C3, A4+B1+B5+C3, A4+B1+B6+C3, A4+B1+B7+C3, A4+B1+B8+C3, A4+B2+B3+C3, A4+B2+B4+C3, A4+B2+B5+C3, A4+B2+B6+C3, A4+B2+B7+C3, A4+B2+B8+C3, A4+B3+B4+C3, A4+B3+B5+C3, A4+B3+B6+C3, A4+B3+B7+C3, A4+B3+B8+C3, A4+B4+B5+C3, A4+B4+B6+C3, A4+B4+B7+C3, A4+B4+B8+C3, A4+B5+B6+C3, A4+B5+B7+C3, A4+B5+B8+C3, A4+B6+B7+C3, A4+B6+B8+C3, A4+B7+B8+C3,
25	A5+B1+B2+C3, A5+B1+B3+C3, A5+B1+B4+C3, A5+B1+B5+C3, A5+B1+B6+C3, A5+B1+B7+C3, A5+B1+B8+C3, A5+B2+B3+C3, A5+B2+B4+C3, A5+B2+B5+C3, A5+B2+B6+C3, A5+B2+B7+C3, A5+B2+B8+C3, A5+B3+B4+C3, A5+B3+B5+C3, A5+B3+B6+C3, A5+B3+B7+C3, A5+B3+B8+C3, A5+B4+B5+C3, A5+B4+B6+C3, A5+B4+B7+C3, A5+B4+B8+C3, A5+B5+B6+C3, A5+B5+B7+C3, A5+B5+B8+C3, A5+B6+B7+C3, A5+B6+B8+C3, A5+B7+B8+C3, A2+A3+B1+C3, A2+A3+B2+C3,
30	A2+A3+B3+C3, A2+A3+B4+C3, A2+A3+B5+C3, A2+A3+B6+C3, A2+A3+B7+C3, A2+A3+B8+C3, A2+A4+B1+C3, A2+A4+B2+C3, A2+A4+B3+C3, A2+A4+B4+C3, A2+A4+B5+C3, A2+A4+B6+C3, A2+A4+B7+C3, A2+A4+B8+C3, A3+A4+B1+C3, A3+A4+B2+C3, A3+A4+B3+C3, A3+A4+B4+C3, A3+A4+B5+C3, A3+A4+B6+C3, A3+A4+B7+C3, A3+A4+B8+C3, A4+A5+B1+C3, A4+A5+B2+C3, A4+A5+B3+C3, A4+A5+B4+C3, A4+A5+B5+C3, A4+A5+B6+C3, A4+A5+B7+C3, A4+A5+B8+C3,
35	A1+A2+A3+C4, A1+A2+A4+C4, A1+A2+A5+C4, A1+A3+A4+C4, A1+A3+A5+C4, A1+A4+A5+C4, A1+A2+B1+C4, A1+A2+B2+C4, A1+A2+B3+C4, A1+A2+B4+C4, A1+A2+B5+C4, A1+A2+B6+C4, A1+A2+B7+C4, A1+A2+B8+C4, A1+A3+B1+C4, A1+A3+B2+C4, A1+A3+B3+C4, A1+A3+B4+C4, A1+A3+B5+C4, A1+A3+B6+C4, A1+A3+B7+C4, A1+A3+B8+C4, A1+A4+B1+C4, A1+A4+B2+C4, A1+A4+B3+C4, A1+A4+B4+C4, A1+A4+B5+C4, A1+A4+B6+C4, A1+A4+B7+C4, A1+A4+B8+C4,
40	A1+A5+B1+C4, A1+A5+B2+C4, A1+A5+B3+C4, A1+A5+B4+C4, A1+A5+B5+C4, A1+A5+B6+C4,

- $A1+A5+B7+C4$, $A1+A5+B8+C4$, $A1+B1+B2+C4$, $A1+B1+B3+C4$, $A1+B1+B4+C4$, $A1+B1+B5+C4$,
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5 $A1+B5+B6+C4$, $A1+B5+B7+C4$, $A1+B5+B8+C4$, $A1+B6+B7+C4$, $A1+B6+B8+C4$, $A1+B7+B8+C4$,
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25 $A2+A3+B3+C4$, $A2+A3+B4+C4$, $A2+A3+B5+C4$, $A2+A3+B6+C4$, $A2+A3+B7+C4$, $A2+A3+B8+C4$,
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30 $A1+A2+A3+C5$, $A1+A2+A4+C5$, $A1+A2+A5+C5$, $A1+A3+A4+C5$, $A1+A3+A5+C5$, $A1+A4+A5+C5$,
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35 $A1+A5+B1+C5$, $A1+A5+B2+C5$, $A1+A5+B3+C5$, $A1+A5+B4+C5$, $A1+A5+B5+C5$, $A1+A5+B6+C5$,
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- A1+B5+B6+C5, A1+B5+B7+C5, A1+B5+B8+C5, A1+B6+B7+C5, A1+B6+B8+C5, A1+B7+B8+C5,
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 25 A4+A5+B3+C5, A4+A5+B4+C5, A4+A5+B5+C5, A4+A5+B6+C5, A4+A5+B7+C5, A4+A5+B8+C5,

 A1+A2+A3+C6, A1+A2+A4+C6, A1+A2+A5+C6, A1+A3+A4+C6, A1+A3+A5+C6, A1+A4+A5+C6,
 A1+A2+B1+C6, A1+A2+B2+C6, A1+A2+B3+C6, A1+A2+B4+C6, A1+A2+B5+C6, A1+A2+B6+C6,
 A1+A2+B7+C6, A1+A2+B8+C6, A1+A3+B1+C6, A1+A3+B2+C6, A1+A3+B3+C6, A1+A3+B4+C6,
 A1+A3+B5+C6, A1+A3+B6+C6, A1+A3+B7+C6, A1+A3+B8+C6, A1+A4+B1+C6, A1+A4+B2+C6,
 30 A1+A4+B3+C6, A1+A4+B4+C6, A1+A4+B5+C6, A1+A4+B6+C6, A1+A4+B7+C6, A1+A4+B8+C6,
 A1+A5+B1+C6, A1+A5+B2+C6, A1+A5+B3+C6, A1+A5+B4+C6, A1+A5+B5+C6, A1+A5+B6+C6,
 A1+A5+B7+C6, A1+A5+B8+C6, A1+B1+B2+C6, A1+B1+B3+C6, A1+B1+B4+C6, A1+B1+B5+C6,
 A1+B1+B6+C6, A1+B1+B7+C6, A1+B1+B8+C6, A1+B2+B3+C6, A1+B2+B4+C6, A1+B2+B5+C6,
 A1+B2+B6+C6, A1+B2+B7+C6, A1+B2+B8+C6, A1+B3+B4+C6, A1+B3+B5+C6, A1+B3+B6+C6,
 35 A1+B3+B7+C6, A1+B3+B8+C6, A1+B4+B5+C6, A1+B4+B6+C6, A1+B4+B7+C6, A1+B4+B8+C6,
 A1+B5+B6+C6, A1+B5+B7+C6, A1+B5+B8+C6, A1+B6+B7+C6, A1+B6+B8+C6, A1+B7+B8+C6,
 A2+B1+B2+C6, A2+B1+B3+C6, A2+B1+B4+C6, A2+B1+B5+C6, A2+B1+B6+C6, A2+B1+B7+C6,
 A2+B1+B8+C6, A2+B2+B3+C6, A2+B2+B4+C6, A2+B2+B5+C6, A2+B2+B6+C6, A2+B2+B7+C6,
 A2+B2+B8+C6, A2+B3+B4+C6, A2+B3+B5+C6, A2+B3+B6+C6, A2+B3+B7+C6, A2+B3+B8+C6,

- A2+B4+B5+C6, A2+B4+B6+C6, A2+B4+B7+C6, A2+B4+B8+C6, A2+B5+B6+C6, A2+B5+B7+C6,
 A2+B5+B8+C6, A2+B6+B7+C6, A2+B6+B8+C6, A2+B7+B8+C6, A3+B1+B2+C6, A3+B1+B3+C6,
 A3+B1+B4+C6, A3+B1+B5+C6, A3+B1+B6+C6, A3+B1+B7+C6, A3+B1+B8+C6, A3+B2+B3+C6,
 A3+B2+B4+C6, A3+B2+B5+C6, A3+B2+B6+C6, A3+B2+B7+C6, A3+B2+B8+C6, A3+B3+B4+C6,
 5 A3+B3+B5+C6, A3+B3+B6+C6, A3+B3+B7+C6, A3+B3+B8+C6, A3+B4+B5+C6, A3+B4+B6+C6,
 A3+B4+B7+C6, A3+B4+B8+C6, A3+B5+B6+C6, A3+B5+B7+C6, A3+B5+B8+C6, A3+B6+B7+C6,
 A3+B6+B8+C6, A3+B7+B8+C6, A4+B1+B2+C6, A4+B1+B3+C6, A4+B1+B4+C6, A4+B1+B5+C6,
 A4+B1+B6+C6, A4+B1+B7+C6, A4+B1+B8+C6, A4+B2+B3+C6, A4+B2+B4+C6, A4+B2+B5+C6,
 A4+B2+B6+C6, A4+B2+B7+C6, A4+B2+B8+C6, A4+B3+B4+C6, A4+B3+B5+C6, A4+B3+B6+C6,
 10 A4+B3+B7+C6, A4+B3+B8+C6, A4+B4+B5+C6, A4+B4+B6+C6, A4+B4+B7+C6, A4+B4+B8+C6,
 A4+B5+B6+C6, A4+B5+B7+C6, A4+B5+B8+C6, A4+B6+B7+C6, A4+B6+B8+C6, A4+B7+B8+C6,
 A5+B1+B2+C6, A5+B1+B3+C6, A5+B1+B4+C6, A5+B1+B5+C6, A5+B1+B6+C6, A5+B1+B7+C6,
 A5+B1+B8+C6, A5+B2+B3+C6, A5+B2+B4+C6, A5+B2+B5+C6, A5+B2+B6+C6, A5+B2+B7+C6,
 A5+B2+B8+C6, A5+B3+B4+C6, A5+B3+B5+C6, A5+B3+B6+C6, A5+B3+B7+C6, A5+B3+B8+C6,
 15 A5+B4+B5+C6, A5+B4+B6+C6, A5+B4+B7+C6, A5+B4+B8+C6, A5+B5+B6+C6, A5+B5+B7+C6,
 A5+B5+B8+C6, A5+B6+B7+C6, A5+B6+B8+C6, A5+B7+B8+C6, A2+A3+B1+C6, A2+A3+B2+C6,
 A2+A3+B3+C6, A2+A3+B4+C6, A2+A3+B5+C6, A2+A3+B6+C6, A2+A3+B7+C6, A2+A3+B8+C6,
 A2+A4+B1+C6, A2+A4+B2+C6, A2+A4+B3+C6, A2+A4+B4+C6, A2+A4+B5+C6, A2+A4+B6+C6,
 A2+A4+B7+C6, A2+A4+B8+C6, A3+A4+B1+C6, A3+A4+B2+C6, A3+A4+B3+C6, A3+A4+B4+C6,
 20 A3+A4+B5+C6, A3+A4+B6+C6, A3+A4+B7+C6, A3+A4+B8+C6, A4+A5+B1+C6, A4+A5+B2+C6,
 A4+A5+B3+C6, A4+A5+B4+C6, A4+A5+B5+C6, A4+A5+B6+C6, A4+A5+B7+C6, A4+A5+B8+C6.

The skilled person will know that there are a number of peptides and proteins known in the
 art that are able to bind to receptors expressed on the luminal side of brain vasculature. The
 sequences or CDRs of these peptides or proteins are non-limiting examples of binding sites that can
 25 be employed in the multispecific binding molecules and are listed in Table 8.

Table 8; peptide- and protein-based BBB shuttles

Proposed target	shuttle	Sequence/CDR	Reference
LRP1	Angiopep-2	TFYQGSRGKKNFKEFY-OH	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
LRP2, LDLR	ApoB (3371-3409)	SSVIDALQYKLEPGTTRITKE- RGLRLATAISLSNKFVEQS	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
LRP1, LRP2, LDLR	ApoE (159-167)	{LRLRLRLI}	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
LDLR	peptide-22	Ac-CR(L)MEPLNGC(R)-NH ₂	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
TfR1	THR	TRKPPWSPVWF-NH ₂	Olla-Salvia et al. Chem. Soc. Rev.

			2016, 45:4690-4707
TfR1	THR <i>retro enantio</i>	<chem>pwvpwmpspch-NH2</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
TfR1,	CRT	<chem>C(S)NTGFSVC(S)</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
Leptin receptors	Leptin30	<chem>YQQLISMPSRNVQSNLLENLADLIHV</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
nAchR	RVG29	<chem>YTWMPENFRIGTFCRFTNSNGKRASNG-OH</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
nAchR	CDX	<chem>GcirtGraerwck-OH</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
KCa Channel	Apamin	<chem>C(S)NC(S)KAPETALC(S)-AR-RC(S)QQH-NH2</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
KCa channel	MiniAP-4	<chem>[Dep](S)KAPETALD(S)</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
GSH transporter	GSH	<chem>γ-L-glutamyl-OH</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
GM1	G23	<chem>HLMLSTWNYEC</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
Unknown	g7	<chem>GFQFES(O-β-Glc)-NH2</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
Unknown	TGN	<chem>TQNYEALHPHNG</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
AMT	TAT(47-57)	<chem>YGREKRNQRR-NH2</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
AMT	SynB1	<chem>RGGRISYSRRKSFSTGR</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
Passive diffusion	(phenylproline)4-NH2	<chem>S(N-MePhe)-(N-MePhe)Diketopiperazines</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
Passive diffusion	Diketopiperazine	<chem>(Phenylproline)4-NH2</chem>	Olla-Salvia et al. Chem. Soc. Rev. 2016, 45:4690-4707
Cdc50A	FC5		
TfR		SYVMH (CDR-1 (HC)) EIAPTNGRTNYIEKPKS (CDR-2 (HC)) GTRAYHY (CDR-3 (HC)) RASDNLYSNLA (CDR-1 (LC)) DATNLAD (CDR-2 (LC)) QHPWGTPLT (CDR-3 (LC)) EVQLVQSGAEVKKPGASVKVSCK ASGYTFTSYWMHWVRQAPGQRLE	US9708406B2

		WIGEIAPTNGRTNYIEKFKSRATLT VDKSASTAYMELSSLRSEDATVYY CARGTRAYHYWGQGMVTVSS (HC) DIQMTQSPSSLSASVGDRVITTCRA SDNLYSNLAWYQQKPGKSPKLLV YDATNLADGVPSRFGSGSGTDYT LTISSLQPEDFATYYCQHFVGWGPLT FGQGTKVEIK (LC)	
TfR	AB405/AB22 I	GFTFSNYGMH MIYYDSSKMNYADTVKG PTSHYVVDV QASQDIGNWLA GATSLAD LQAYNTPWT	WO 2015191934 A2
Basigin	Anti-BsgA	EVQLVESGGGLVLPGRSMKLSCAA <u>SGFTFRTYYMAWVRQAPTKGLEW</u> VASISIGDNTYYRDSVMGRFTISR DDAKSTLHLQMDLRSEDATYYC <u>VRLRGYFDYWGQGMVTVSS</u> (HC) EIVLTQSPATMPASPGEKVTLTCRA <u>SSSIRYIYWYQQKSGTSPKLWTFD</u> <u>TSKLASGVPRFSGSGSGTSYSLTIS</u> METEDTATYYCQOGRSYPLTFGSG TKLEIK (LC)	WO 2016094566 A2
Basigin	Anti-BsgB	EVQLVESGGGLVQPGRSLKLSCTVA <u>SGFTFNYYWMTWIRQAPGKGLEW</u> <u>FASITNTGGSTYYPDVSKGRFTISR</u> DNAQSTLYLQTNLRPEDTATYYC <u>ARRDGSYYPIYWFYDLWGPGTT</u> VTVSS (HC) NTVMTQSPTSMFISVGDRVMTNCK <u>ASRSVGTNVDWYQQKGTGQSPILLF</u> <u>YGASRYIGVPDRFTGSGSGTDFTLT</u> ISMQAEDLAVYYCLOYNYNWAFG GGTKLEIK (LC)	WO 2016094566 A2
Basigin	Anti-BsgC	EVQLVESGGSLVQPGRSMKVSCAA <u>SGFTFTKYYMAWVRQAPTKGLEW</u> VASISTGGGNTYYRDSVKGRTISR DNAKSTLYLQMDSLRSEDATYYC	WO 2016094566 A2

		<u>ARTLINYS</u> <u>ADYVMD</u> <u>AWGQ</u> <u>GAS</u> VTVSS (HC) <u>DIQMTQSPASLSASL</u> <u>GETV</u> <u>SIECL</u> <u>A</u> <u>SEGISNSLAWYQQKPGKSPQLLIYG</u> <u>ASSLQDGVPSRFS</u> <u>SGSG</u> <u>SGTQFSLKI</u> <u>SGMQPEDEGIYYCQQGYKYPFTFG</u> SGTKEIK (LC)	
Basigin	Anti-BsgD	EVQLVESGGGLVQPGSRMKLSCA <u>ASGFTLSNYYMAWVRQ</u> <u>APTKGLE</u> <u>WVASISTGGGYTYRDSVKGRFTI</u> <u>SRDLAKSTLYLQMDSLRSEDTATY</u> <u>HCARSLIN</u> <u>YRNYGDYVMD</u> <u>AWGQ</u> GASVTVSS (HC) <u>DIQMTQSPASLSASL</u> <u>GETV</u> <u>SIECL</u> <u>A</u> <u>SEGISNSLAWYQQKPGKSPQLLIYD</u> <u>ASSLQVGVPSRFS</u> <u>SGSG</u> <u>SGTQYSLKI</u> <u>SGLQPEDEGVYYCQQGYKYPFTFG</u> SGTKEIK (LC)	WO 2016094566 A2
Basiging	Anti-BsgE	QVQLKESGPGLVQPSQTLSTCSVS <u>GLSLTSSLSWIRQPPGKGLEWMG</u> <u>GIWSKGGTEYN</u> <u>SPIKSRLSIS</u> <u>RDTSK</u> <u>SQIFLKMNSLQTEDTAMYFC</u> <u>ARNG</u> <u>VYHNYWYFDEWGP</u> <u>GMTVTVSS</u> (HC) <u>QFTLTQPKSVSGSLRSTITIP</u> <u>CERSS</u> <u>GDIGHNYVSWYQQHLGRPPINVIY</u> <u>ADDQRPSEVSDRFS</u> <u>GSIDSSNSAS</u> <u>LTITNLQMDDEADYFCQSYDSNVD</u> <u>IVFGG</u> <u>GKLT</u> <u>TVL</u> (LC)	WO 2016094566 A2
GlutI	Anti-GlutI	QVQLQQPGSVLVRPGASVKLSCKA <u>SGYTFTGSWLHWAKQRPGQGLEW</u> <u>IGEIH</u> <u>PYSGNTNYNERFKG</u> <u>KATLTV</u> <u>DTPSS</u> <u>TAYVDLRSLTFEDSAVYYC</u> <u>AKEGGWFLRIYGMDYWGQ</u> <u>GTSVT</u> VSS (HC) <u>DIVLTQSPSSLSASLGD</u> <u>TITITCHAS</u> <u>QNINVWLSWYQQKPGNIPKLLIYK</u> <u>ASNLHSGVPSRFS</u> <u>SGSG</u> <u>SGTGFTLTIS</u> <u>SLQPEDIATYYCQQGQTFPYTFGG</u> GTRLEIK (LC)	WO 2016094566 A2

Therapeutic use

In one embodiment of the invention the multispecific binding molecules are used for the diagnosis, prevention or treatment of neurodegenerative disorders, selected from the group comprising AD, CAA, CTE, MSA, LBD, CBD, PSP, HD, ALS, FTD, FTDP-17 and PD.

- 5 In particular embodiments, the antibodies or antigen-binding fragments thereof disclosed herein may be used alone, or in combination with therapeutics known in the art for the treatment of NDDs.

Methods of Making Antibodies and Antigen-binding Fragments Thereof

- 10 The present invention includes methods for making multispecific antibodies or antigen-binding fragments thereof. The skilled person will know that there are a variety of methods that allow the production of multispecific binding molecules such as multispecific antibodies. See Brinkmann, U., Kontermann, R.E, MAbs, 2017, 9 (2), 182-212.

- 15 Methods for producing and screening for monospecific antibodies using hybridoma technology are routine and well known in the art. In a non-limiting example, mice can be immunized with an antigen of interest or a cell expressing such an antigen. Once an immune response is detected, e.g., antibodies specific for the antigen are detected in the mouse serum, the mouse spleen is harvested and splenocytes isolated. B-cells are cultured, as described by Steenbakkers et al., 1994, Mol. Biol. Rep. 19: 125-134.

- 20 B-cell clones from reactive supernatants are then immortalized, e.g. by mini-electrofusion following published procedures (Steenbakkers et al., J. Immunol. Meth. 1992, 152: 69-77; Steenbakkers et al., 1994, Mol. Biol. Rep. 19:125-34). Hybridomas are selected and cloned by limiting dilution.

- 25 The hybridoma clones are then assayed by methods known in the art for cells that secrete antibodies capable of binding the antigen. Ascites fluid, which generally contains high levels of antibodies, can be generated by inoculating mice intraperitoneally with positive hybridoma clones.

- 30 Monoclonal antibodies derived from animals other than rats and mice offer unique advantages. Many protein targets relevant to signal transduction and disease are highly conserved between mice, rats and humans, and can therefore be recognized as self-antigens by a mouse or rat host, making them less immunogenic. This problem may be avoided when using rabbit as a host animal. See, e.g., Rossi et al., Am. J. Clin. Pathol., 2005, 124, 295-302.

Adjuvants that can be used in the methods of antibody generation include, but are not limited to, protein adjuvants; bacterial adjuvants, e.g., whole bacteria (BCG, *Corynebacterium parvum*, *Salmonella minnesota*) and bacterial components including cell wall skeleton, trehalose dimycolate, monophosphoryl lipid A, methanol extractable residue (MER) of tubercle bacillus,

complete or incomplete Freund's adjuvant; viral adjuvants; chemical adjuvants, e.g., aluminum hydroxide, iodoacetate and cholesteryl hemisuccinate; naked DNA adjuvants. Other adjuvants that can be used in the methods of the invention include, Cholera toxin, paropox proteins, MF-59 (Chiron Corporation; See also Bieg et al. (1999) "GAD65 And Insulin B Chain Peptide (9-23) Are Not Primary Autoantigens In The Type 1 Diabetes Syndrome Of The BB Rat," *Autoimmunity*, 31(1):15-24, which is incorporated herein by reference), MPL® (Corixa Corporation; See also Lodmell et al. (2000) "DNA Vaccination Of Mice Against Rabies Virus: Effects Of The Route Of Vaccination And The Adjuvant Monophosphoryl Lipid A (MPL)," *Vaccine*, 18: 1059-1066; Johnson et al. (1999) "3-O-Desacyl Monophosphoryl Lipid A Derivatives: Synthesis And Immunostimulant Activities," *Journal of Medicinal Chemistry*, 42: 4640-4649; Baldrige et al. (1999) "Monophosphoryl Lipid A (MPL) Formulations For The Next Generation Of Vaccines," *Methods*, 19: 103-107, all of which are incorporated herein by reference), RC-529 adjuvant (Corixa Corporation; the lead compound from Corixa's aminoalkyl glucosaminide 4-phosphate (AGP) chemical library, see also www.corixa.com), and DETOX™ adjuvant (Corixa Corporation; DETOX™ adjuvant includes MPL® adjuvant (monophosphoryl lipid A) and mycobacterial cell wall skeleton; See also Eton et al. (1998) "Active Immunotherapy With Ultraviolet B-Irradiated Autologous Whole Melanoma Cells Plus DETOX In Patients With Metastatic Melanoma," *Clin. Cancer Res.* 4(3):619-627; and Gupta et al. (1995) "Adjuvants For Human Vaccines—Current Status, Problems And Future Prospects," *Vaccine*, 13(14): 1263-1276, both of which are incorporated herein by reference).

Antibodies can also be produced using transgenic mice which are incapable of expressing functional endogenous immunoglobulins, but which can express human immunoglobulin genes. For example, the human heavy and light chain immunoglobulin gene complexes may be introduced randomly or by homologous recombination into mouse embryonic stem cells. Alternatively, the human variable region, constant region, and diversity region may be introduced into mouse embryonic stem cells in addition to the human heavy and light chain genes. The mouse heavy and light chain immunoglobulin genes may be rendered non-functional separately or simultaneously with the introduction of human immunoglobulin loci by homologous recombination. In particular, homozygous deletion of the JH region prevents endogenous antibody production. The modified embryonic stem cells are expanded and microinjected into blastocysts to produce chimeric mice. The chimeric mice are then bred to produce homozygous offspring which express human antibodies. The transgenic mice are immunized using conventional methodologies with a selected antigen, e.g., all or a portion of a polypeptide of the invention. Monoclonal antibodies directed against the antigen can be obtained from the immunized, transgenic mice using conventional hybridoma technology. The human immunoglobulin transgenes harbored by the transgenic mice rearrange during B cell differentiation, and subsequently undergo class switching and somatic mutation. Thus, using such a technique, it is possible to produce therapeutically useful IgG, IgA, IgM and IgE antibodies. For an overview of this technology for producing human antibodies, see Lonberg et al. (1995) "Human Antibodies From Transgenic Mice," *Int. Rev. Immunol.* 13:65-93, which is incorporated herein by reference in its entirety. For a detailed discussion of this technology for producing human antibodies and human

monoclonal antibodies and protocols for producing such antibodies, see, e.g., International Publication Nos. WO 98/24893, WO 96/34096, and WO 96/33735; and U.S. Pat. Nos. 5,413,923, 5,625,126, 5,633,425, 5,569,825, 5,661,016, 5,545,806, 5,814,318, and 5,939,598, which are incorporated by reference herein in their entirety. In addition, companies such as Abgenix/Amgen(Freemont, Calif.) and Medarex/BMS (Princeton, N.J.), Kymab (Cambridge, UK) and Merus (Utrecht, Netherlands) can be engaged to provide human antibodies directed against a selected antigen using technology similar to that described above.

The antibodies disclosed herein may also be produced recombinantly (e.g., in an *E. coli*/T7 expression system, a mammalian cell expression system or a lower eukaryote expression system). In this embodiment, nucleic acids encoding the antibody immunoglobulin molecules of the invention (e.g., V_H or V_L) may be inserted into a pET-based plasmid and expressed in the *E. coli*/T7 system. For example, the present invention includes methods for expressing an antibody or antigen-binding fragment thereof or immunoglobulin chain thereof in a host cell (e.g., bacterial host cell such as *E.coli* such as BL21 or BL21DE3) comprising expressing T7 RNA polymerase in the cell which also includes a polynucleotide encoding an immunoglobulin chain that is operably linked to a T7 promoter. For example, in an embodiment of the invention, a bacterial host cell, such as a *E. coli*, includes a polynucleotide encoding the T7 RNA polymerase gene operably linked to a *lac* promoter and expression of the polymerase and the chain is induced by incubation of the host cell with IPTG (isopropyl-beta-D-thiogalactopyranoside).

Monoclonal antibody preparations can be produced using a wide variety of techniques known in the art including the use of hybridoma, recombinant, and phage display technologies, or a combination thereof. For example, monoclonal antibodies can be produced using hybridoma techniques including those known in the art and taught, for example, in Harlow et al., ANTIBODIES: A LABORATORY MANUAL, (Cold Spring Harbor Laboratory Press, 2nd ed. 1988); Hammerling, et al., in: MONOCLONAL ANTIBODIES AND T-CELL HYBRIDOMAS, pp. 563-681 (Elsevier, N.Y., 1981) (both of which are incorporated by reference in their entireties). The term "monoclonal antibody" as used herein is not limited to antibodies produced through hybridoma technology. The term "monoclonal antibody" refers to an antibody that is derived from a single clone, including any eukaryotic, prokaryotic, or phage clone, and not the method by which it is produced. One example of a method for recombinant production of antibodies is disclosed in U.S. Patent No. 4,816,567.

Thus, the present invention includes recombinant methods for making an antibody or antigen-binding fragment thereof of the present invention, or an immunoglobulin chain thereof, comprising introducing a polynucleotide encoding one or more immunoglobulin chains of the antibody or fragment (e.g., heavy and/or light immunoglobulin chain); culturing the host cell (e.g., CHO or *Pichia* or *Pichia pastoris*) under condition favorable to such expression and, optionally, isolating the antibody or fragment or chain from the host cell and/or medium in which the host cell is grown.

Antibodies of the present invention can also be synthesized by any of the methods set forth in U.S. Patent No. 6,331,415.

Eukaryotic and prokaryotic host cells, including mammalian cells as hosts for expression of the antibodies or fragments or immunoglobulin chains disclosed herein are well known in the art and include many immortalized cell lines available from the American Type Culture Collection (ATCC). These include, *inter alia*, Chinese hamster ovary (CHO) cells, NSO, SP2 cells, HeLa cells, baby hamster kidney (BHK) cells, monkey kidney cells (COS), human hepatocellular carcinoma cells (e.g., Hep G2), A549 cells, 3T3 cells, HEK-293 cells and a number of other cell lines. Mammalian host cells include human, mouse, rat, dog, monkey, pig, goat, bovine, horse and hamster cells. Cell lines of particular preference are selected through determining which cell lines have high expression levels. Other cell lines that may be used are insect cell lines, such as Sf9 cells, amphibian cells, bacterial cells, plant cells and fungal cells. Fungal cells include yeast and filamentous fungus cells including, for example, *Pichia pastoris*, *Pichia finlandica*, *Pichia trehalophila*, *Pichia kocklamae*, *Pichia membranaefaciens*, *Pichia minuta* (*Ogataea minuta*, *Pichia lindneri*), *Pichia opuntiae*, *Pichia thermotolerans*, *Pichia salictaria*, *Pichia guercuum*, *Pichia pipperi*, *Pichia stiptis*, *Pichia methanolica*, *Pichia* sp., *Saccharomyces cerevisiae*, *Saccharomyces* sp., *Hansenula polymorpha*, *Kluyveromyces* sp., *Kluyveromyces lactis*, *Candida albicans*, *Aspergillus nidulans*, *Aspergillus niger*, *Aspergillus oryzae*, *Trichoderma reesei*, *Chrysosporium lucknowense*, *Fusarium* sp., *Fusarium gramineum*, *Fusarium venenatum*, *Physcomitrella patens* and *Neurospora crassa*. *Pichia* sp., any *Saccharomyces* sp., *Hansenula polymorpha*, any *Kluyveromyces* sp., *Candida albicans*, any *Aspergillus* sp., *Trichoderma reesei*, *Chrysosporium lucknowense*, any *Fusarium* sp., *Yarrowia lipolytica*, and *Neurospora crassa*. When recombinant expression vectors encoding the heavy chain or antigen-binding portion or fragment thereof, the light chain and/or antigen-binding fragment thereof are introduced into mammalian host cells, the antibodies are produced by culturing the host cells for a period of time sufficient to allow for expression of the antibody or fragment or chain in the host cells or secretion of the into the culture medium in which the host cells are grown.

A variety of host-expression vector systems may be utilized to express the antibodies of the invention. Such host-expression systems represent vehicles by which the coding sequences of the antibodies may be produced and subsequently purified, but also represent cells which may, when transformed or transfected with the appropriate nucleotide coding sequences, express the antibodies of the invention in situ. These include, but are not limited to, microorganisms such as bacteria (e.g., *E. coli* and *B. subtilis*) transformed with recombinant bacteriophage DNA, plasmid DNA or cosmid DNA expression vectors containing immunoglobulin coding sequences; yeast (e.g., *Saccharomyces*, *Pichia*) transformed with recombinant yeast expression vectors containing immunoglobulin coding sequences; insect cell systems infected with recombinant virus expression vectors (e.g., baculovirus) containing the immunoglobulin coding sequences; plant cell systems infected with recombinant virus expression vectors (e.g., cauliflower mosaic virus (CaMV) and tobacco mosaic virus (TMV)) or transformed with recombinant plasmid expression vectors (e.g., Ti plasmid) containing immunoglobulin coding sequences; or mammalian cell systems (e.g., COS, CHO, BHK, 293, 293T, 3T3

cells, lymphotic cells (see U.S. Pat. No. 5,807,715), Per C.6 cells (rat retinal cells developed by Crucell) harboring recombinant expression constructs containing promoters derived from the genome of mammalian cells (e.g., metallothionein promoter) or from mammalian viruses (e.g., the adenovirus late promoter; the vaccinia virus 7.5K promoter).

5 In bacterial systems, a number of expression vectors may be advantageously selected depending upon the use intended for the antibody being expressed. For example, when a large quantity of such a protein is to be produced, for the generation of pharmaceutical compositions of an antibody, vectors which direct the expression of high levels of fusion protein products that are readily purified may be desirable. Such vectors include, but are not limited, to the E. coli expression vector
10 pUR278 (Ruther et al. (1983) "Easy Identification Of cDNA Clones," EMBO J. 2:1791-1794), in which the antibody coding sequence may be ligated individually into the vector in frame with the lac Z coding region so that a fusion protein is produced; pIN vectors (Inouye et al. (1985) "Up-Promoter Mutations In The Lpp Gene Of Escherichia coli," Nucleic Acids Res. 13:3101-3110; Van Heeke et al. (1989) "Expression Of Human Asparagine Synthetase In Escherichia coli," J. Biol. Chem. 264:5503-
15 5509); and the like. pGEX vectors may also be used to express foreign polypeptides as fusion proteins with glutathione S-transferase (GST). In general, such fusion proteins are soluble and can easily be purified from lysed cells by adsorption and binding to a matrix glutathione-agarose beads followed by elution in the presence of free glutathione. The pGEX vectors are designed to include thrombin or factor Xa protease cleavage sites so that the cloned target gene product can be released from the GST
20 moiety.

 In an insect system, *Autographa californica* nuclear polyhedrosis virus (AcNPV) is used as a vector to express foreign genes. The virus grows in *Spodoptera frugiperda* cells. The antibody coding sequence may be cloned individually into non-essential regions (e.g., the polyhedrin gene) of the virus and placed under control of an AcNPV promoter (e.g., the polyhedrin promoter).

25 In mammalian host cells, a number of viral-based expression systems may be utilized. In cases where an adenovirus is used as an expression vector, the antibody coding sequence of interest may be ligated to an adenovirus transcription/translation control complex, e.g., the late promoter and tripartite leader sequence. This chimeric gene may then be inserted in the adenovirus genome by in vitro or in vivo recombination. Insertion in a non-essential region of the viral genome (e.g., region E1
30 or E3) will result in a recombinant virus that is viable and capable of expressing the immunoglobulin molecule in infected hosts. (see e.g., see Logan et al. (1984) "Adenovirus Tripartite Leader Sequence Enhances Translation Of mRNAs Late After Infection," Proc. Natl. Acad. Sci. (U.S.A.) 81:3655-3659). Specific initiation signals may also be required for efficient translation of inserted antibody coding sequences. These signals include the ATG initiation codon and adjacent sequences. Furthermore, the
35 initiation codon must be in phase with the reading frame of the desired coding sequence to ensure translation of the entire insert. These exogenous translational control signals and initiation codons can be of a variety of origins, both natural and synthetic. The efficiency of expression may be enhanced by the inclusion of appropriate transcription enhancer elements, transcription terminators,

etc. (see Bitter et al. (1987) "Expression And Secretion Vectors For Yeast," *Methods in Enzymol.* 153:516-544).

In addition, a host cell strain may be chosen which modulates the expression of the inserted sequences, or modifies and processes the gene product in the specific fashion desired. Such
5 modifications (e.g., glycosylation) and processing (e.g., cleavage) of protein products may be important for the function of the protein. Different host cells have characteristic and specific mechanisms for the post-translational processing and modification of proteins and gene products. Appropriate cell lines or host systems can be chosen to ensure the correct modification and processing of the foreign protein expressed. To this end, eukaryotic host cells which possess the cellular machinery for proper
10 processing of the primary transcript, glycosylation, and phosphorylation of the gene product may be used. Such mammalian host cells include but are not limited to CHO, VERY, BHK, HeLa, COS, MDCK, 293, 293T, 3T3, WI38, BT483, Hs578T, HTB2, BT20 and T47D, CRL7030 and Hs578Bst.

For long-term, high-yield production of recombinant proteins, stable expression is preferred. For example, cell lines which stably express an antibody of the invention may be
15 engineered. Rather than using expression vectors which contain viral origins of replication, host cells can be transformed with DNA controlled by appropriate expression control elements (e.g., promoter, enhancer, sequences, transcription terminators, polyadenylation sites, etc.), and a selectable marker. Following the introduction of the foreign DNA, engineered cells may be allowed to grow for 1-2 days in an enriched media, and then are switched to a selective media. The selectable marker in the
20 recombinant plasmid confers resistance to the selection and allows cells to stably integrate the plasmid into their chromosomes and grow to form foci which in turn can be cloned and expanded into cell lines. This method may advantageously be used to engineer cell lines which express the antibodies of the invention. Such engineered cell lines may be particularly useful in screening and evaluation of compounds that interact directly or indirectly with the antibodies of the invention.

25 A number of selection systems may be used, including but not limited to the herpes simplex virus thymidine kinase (Wigler et al. (1977) "Transfer Of Purified Herpes Virus Thymidine Kinase Gene To Cultured Mouse Cells," *Cell* 11:223-232), hypoxanthine-guanine phosphoribosyltransferase (Szybalska et al. (1962) "Genetics Of Human Cess Line. IV. DNA-Mediated Heritable Transformation Of A Biochemical Trait," *Proc. Natl. Acad. Sci. (U.S.A.)* 48:2026-2034), and
30 adenine phosphoribosyltransferase (Lowy et al. (1980) "Isolation Of Transforming DNA: Cloning The Hamster Aprt Gene," *Cell* 22:817-823) genes can be employed in tk⁻, hgp⁻ or aprt⁻ cells, respectively. Also, antimetabolite resistance can be used as the basis of selection for the following genes: dhfr, which confers resistance to methotrexate (Wigler et al. (1980) "Transformation Of Mammalian Cells With An Amplifiable Dominant-Acting Gene," *Proc. Natl. Acad. Sci. (U.S.A.)* 77:3567-3570; O'Hare et al. (1981) "Transformation Of Mouse Fibroblasts To Methotrexate Resistance By A Recombinant Plasmid Expressing A Prokaryotic Dihydrofolate Reductase," *Proc. Natl. Acad. Sci. (U.S.A.)* 78:1527-1531); gpt, which confers resistance to mycophenolic acid (Mulligan et al. (1981) "Selection For Animal Cells That Express The Escherichia coli Gene Coding For Xanthine-Guanine
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Phosphoribosyltransferase," *Proc. Natl. Acad. Sci. (U.S.A.)* 78:2072-2076); neo, which confers resistance to the aminoglycoside G-418 (Tachibana et al. (1991) "Altered Reactivity Of Immunoglobulin Produced By Human-Human Hybridoma Cells Transfected By pSV2-Neo Gene," *Cytotechnology* 6(3):219-226; Tolstoshev (1993) "Gene Therapy, Concepts, Current Trials And Future Directions," *Ann. Rev. Pharmacol. Toxicol.* 32:573-596; Mulligan (1993) "The Basic Science Of Gene Therapy," *Science* 260:926-932; and Morgan et al. (1993) "Human gene therapy," *Ann. Rev. Biochem.* 62:191-217). Methods commonly known in the art of recombinant DNA technology which can be used are described in Ausubel et al. (eds.), 1993, *CURRENT PROTOCOLS IN MOLECULAR BIOLOGY*, John Wiley & Sons, NY; Kriegler, 1990, *GENE TRANSFER AND EXPRESSION, A LABORATORY MANUAL*, Stockton Press, NY; and in Chapters 12 and 13, Dracopoli et al. (eds), 1994, *CURRENT PROTOCOLS IN HUMAN GENETICS*, John Wiley & Sons, NY.; Colbere-Garapin et al. (1981) "A New Dominant Hybrid Selective Marker For Higher Eukaryotic Cells," *J. Mol. Biol.* 150:1-14; and hygromycin, which confers resistance to hygromycin (Santerre et al. (1984) "Expression Of Prokaryotic Genes For Hygromycin B And G418 Resistance As Dominant-Selection Markers In Mouse L Cells," *Gene* 30:147-156).

The expression levels of an antibody of the invention can be increased by vector amplification (for a review, see Bebbington and Hentschel, "The Use Of Vectors Based On Gene Amplification For The Expression Of Cloned Genes In Mammalian Cells," in *DNA CLONING*, Vol. 3. (Academic Press, New York, 1987)). When a marker in the vector system expressing an antibody is amplifiable, increase in the level of inhibitor present in culture of host cell will increase the number of copies of the marker gene. Since the amplified region is associated with the nucleotide sequence of the antibody, production of the antibody will also increase (Crouse et al. (1983) "Expression And Amplification Of Engineered Mouse Dihydrofolate Reductase Minigenes," *Mol. Cell. Biol.* 3:257-266).

The host cell may be co-transfected with two expression vectors of the invention, the first vector encoding a heavy chain derived polypeptide and the second vector encoding a light chain derived polypeptide. The two vectors may contain identical selectable markers which enable equal expression of heavy and light chain polypeptides. Alternatively, a single vector may be used which encodes both heavy and light chain polypeptides. In such situations, the light chain should be placed before the heavy chain to avoid an excess of toxic free heavy chain (Proudfoot (1986) "Expression And Amplification Of Engineered Mouse Dihydrofolate Reductase Minigenes," *Nature* 322:562-565; Kohler (1980) "Immunoglobulin Chain Loss In Hybridoma Lines," *Proc. Natl. Acad. Sci. (U.S.A.)* 77:2197-2199). The coding sequences for the heavy and light chains may comprise cDNA or genomic DNA.

Antibodies and antigen-binding fragments thereof and immunoglobulin chains can be recovered from the culture medium using standard protein purification methods. Further, expression of antibodies and antigen-binding fragments thereof and immunoglobulin chains of the invention (or other moieties therefrom) from production cell lines can be enhanced using a number of known techniques. For example, the glutamine synthetase gene expression system (the GS system) is a common approach for enhancing expression under certain conditions. The GS system is discussed in

whole or part in connection with European Patent Nos. 0 216 846, 0 256 055, and 0 323 997 and European Patent Application No. 89303964.4. Thus, in an embodiment of the invention, the mammalian host cells (*e.g.*, CHO) lack a glutamine synthetase gene and are grown in the absence of glutamine in the medium wherein, however, the polynucleotide encoding the immunoglobulin chain
5 comprises a glutamine synthetase gene which complements the lack of the gene in the host cell.

The present invention includes methods for purifying an antibody or antigen-binding fragment thereof of the present invention comprising introducing a sample comprising the antibody or fragment to a purification medium (*e.g.*, cation exchange medium, anion exchange medium, hydrophobic exchange medium, affinity purification medium (*e.g.*, protein-A, protein-G, protein-A/G, protein-L)) and either collecting purified antibody or fragment from the flow-through fraction of said
10 sample that does not bind to the medium; or, discarding the flow-through fraction and eluting bound antibody or fragment from the medium and collecting the eluate. In an embodiment of the invention, the medium is in a column to which the sample is applied. In an embodiment of the invention, the purification method is conducted following recombinant expression of the antibody or fragment in a
15 host cell, *e.g.*, wherein the host cell is first lysed and, optionally, the lysate is purified of insoluble materials prior to purification on a medium.

In general, glycoproteins produced in a particular cell line or transgenic animal will have a glycosylation pattern that is characteristic for glycoproteins produced in the cell line or transgenic animal. Therefore, the particular glycosylation pattern of an antibody will depend on the particular
20 cell line or transgenic animal used to produce the antibody. However, all antibodies encoded by the nucleic acid molecules provided herein, or comprising the amino acid sequences provided herein, comprise the instant invention, independent of the glycosylation pattern the antibodies may have. Similarly, in particular embodiments, antibodies with a glycosylation pattern comprising only non-fucosylated *N*-glycans may be advantageous, because these antibodies have been shown to typically
25 exhibit more potent efficacy than their fucosylated counterparts both *in vitro* and *in vivo* (See for example, Shinkawa *et al.*, *J. Biol. Chem.* 278: 3466-3473 (2003); U.S. Patent Nos. 6,946,292 and 7,214,775). These antibodies with non-fucosylated *N*-glycans are not likely to be immunogenic because their carbohydrate structures are a normal component of the population that exists in human serum IgG.

30 The present invention further includes antigen-binding fragments of the antibodies disclosed herein. The antibody fragments include F(ab)₂ fragments, which may be produced by enzymatic cleavage of an IgG by, for example, pepsin. Fab fragments may be produced by, for example, reduction of F(ab)₂ with dithiothreitol or mercaptoethylamine.

Immunoglobulins may be assigned to different classes depending on the amino acid
35 sequences of the constant domain of their heavy chains. In some embodiments, different constant domains may be appended to humanized V_L and V_H regions derived from the CDRs provided herein. There are at least five major classes of immunoglobulins: IgA, IgD, IgE, IgG and IgM, and several of

these may be further divided into subclasses (isotypes), *e.g.* IgG1, IgG2, IgG3 and IgG4; IgA1 and IgA2. The invention comprises antibodies and antigen-binding fragments of any of these classes or subclasses of antibodies.

5 In one embodiment, the antibody or antigen-binding fragment comprises a heavy chain constant region, *e.g.* a human constant region, such as κ 1, κ 2, κ 3, or κ 4 human heavy chain constant region or a variant thereof. In another embodiment, the antibody or antigen-binding fragment comprises a light chain constant region, *e.g.* a human light chain constant region, such as lambda or kappa human light chain region or variant thereof. By way of example, and not limitation the human heavy chain constant region can be κ 4 and the human light chain constant region can be kappa. In an
10 alternative embodiment, the Fc region of the antibody is κ 4 with a Ser228Pro mutation (*Angal S. et al., 1993, Mol Immunol. 30: 105-108* position 241 is based on the Kabat numbering system).

In one embodiment, the antibody or antigen-binding fragment comprises a heavy chain constant region of the IgG1 subtype. In one embodiment, the antibody or antigen-binding fragment comprises a heavy chain constant region of the IgG2 subtype. In one embodiment, the antibody or
15 antigen-binding fragment comprises a heavy chain constant region of the IgG4 subtype.

Experimental and Diagnostic Uses

Multispecific antibodies (*e.g.*, humanized antibodies) and antigen-binding fragments thereof may also be useful in diagnostic assays for targets involved in NDD as discussed herein, *e.g.*, detecting its expression in specific cells, tissues, serum or CSF. Such diagnostic methods may be
20 useful in various disease diagnoses.

The present invention includes ELISA assays (enzyme-linked immunosorbent assay) incorporating the use of a multispecific antibody or antigen-binding fragment thereof disclosed herein.

For example, such a method comprises the following steps:

- (a) coat a substrate (*e.g.*, surface of a microtiter plate well, *e.g.*, a plastic plate) with a multispecific
25 antibody or antigen-binding fragment thereof;
- (b) apply a sample to be tested for the presence of a pathological protein involved in any NDD described herein to the substrate;
- (c) wash the plate, so that unbound material in the sample is removed;
- (d) apply detectably labeled antibodies (*e.g.*, enzyme-linked antibodies) which are also specific to the
30 antigen;
- (e) wash the substrate, so that the unbound, labeled antibodies are removed;
- (f) if the labeled antibodies are enzyme linked, apply a chemical which is converted by the enzyme into a fluorescent signal; and
- (g) detect the presence of the labeled antibody.

Detection of the label associated with the substrate indicates the presence of the pathological protein.

In a further embodiment, the labeled antibody or antigen-binding fragment thereof is labeled with peroxidase which react with ABTS (*e.g.*, 2,2'-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid)) or 3,3',5,5'-Tetramethylbenzidine to produce a color change which is detectable. Alternatively,
5 the labeled antibody or fragment is labeled with a detectable radioisotope (*e.g.*, ^3H) which can be detected by scintillation counter in the presence of a scintillant.

A multispecific antibody or antigen-binding fragment thereof of the invention may be used in a Western blot or immune-protein blot procedure. Such a procedure forms part of the present
10 invention and includes *e.g.*:

(1) optionally transferring proteins from a sample to be tested for the presence of pathological protein (*e.g.*, from a PAGE or SDS-PAGE electrophoretic separation of the proteins in the sample) onto a membrane or other solid substrate using a method known in the art (*e.g.*, semi-dry blotting or tank blotting); contacting the membrane or other solid substrate to be tested for the presence of bound
15 target or a fragment thereof with a multispecific antibody or antigen-binding fragment thereof of the invention.

Such a membrane may take the form of a nitrocellulose or vinyl-based (*e.g.*, polyvinylidene fluoride (PVDF)) membrane to which the proteins to be tested for the presence of pathological protein in a non-denaturing PAGE (polyacrylamide gel electrophoresis) gel or SDS-PAGE (sodium dodecyl
20 sulfate polyacrylamide gel electrophoresis) gel have been transferred (*e.g.*, following electrophoretic separation in the gel). Before contacting the membrane with the multispecific antibody or fragment, the membrane is optionally blocked, *e.g.*, with non-fat dry milk or the like so as to bind non-specific protein binding sites on the membrane.

(2) washing the membrane one or more times to remove unbound multispecific antibody or fragment
25 and other unbound substances; and

(3) detecting the bound multispecific antibody or fragment.

Detection of the bound antibody or fragment indicates that the pathological protein is present on the membrane or substrate and in the sample. Detection of the bound antibody or fragment may be by binding the antibody or fragment with a secondary antibody (an anti-immunoglobulin antibody) which is detectably labeled and, then, detecting the presence of the
30 secondary antibody.

The multispecific antibodies and antigen-binding fragments thereof disclosed herein may also be used for immunohistochemistry. Such a method forms part of the present invention and comprises, *e.g.*,

(1) contacting a cell to be tested for the presence of the pathological protein with a multispecific antibody or antigen-binding fragment thereof of the invention; and

(2) detecting the antibody or fragment on or in the cell.

If the antibody or fragment itself is detectably labeled, it can be detected directly.

5 Alternatively, the antibody or fragment may be bound by a detectably labeled secondary antibody which is detected.

Pharmaceutical Compositions and Administration

To prepare pharmaceutical or sterile compositions of the multispecific antibodies and antigen-binding fragments of the invention, the antibody or antigen-binding fragment thereof is
10 admixed with a pharmaceutically acceptable carrier or excipient. See, *e.g.*, *Remington's Pharmaceutical Sciences* and *U.S. Pharmacopeia: National Formulary*, Mack Publishing Company, Easton, PA (1984). Hence, also included in the present invention are pharmaceutical preparations comprising one of the previously described multispecific binding molecules and a pharmaceutically acceptable carrier or excipient.

15 Formulations of therapeutic and diagnostic agents may be prepared by mixing with acceptable carriers, excipients, or stabilizers in the form of, *e.g.*, lyophilized powders, slurries, aqueous solutions or suspensions (see, *e.g.*, Hardman, *et al.* (2001) *Goodman and Gilman's The Pharmacological Basis of Therapeutics*, McGraw-Hill, New York, NY; Gennaro (2000) *Remington: The Science and Practice of Pharmacy*, Lippincott, Williams, and Wilkins, New York, NY; Avis, *et al.* (eds.)
20 (1993) *Pharmaceutical Dosage Forms: Parenteral Medications*, Marcel Dekker, NY; Lieberman, *et al.* (eds.) (1990) *Pharmaceutical Dosage Forms: Tablets*, Marcel Dekker, NY; Lieberman, *et al.* (eds.) (1990) *Pharmaceutical Dosage Forms: Disperse Systems*, Marcel Dekker, NY; Weiner and Kotkoskie (2000) *Excipient Toxicity and Safety*, Marcel Dekker, Inc., New York, NY).

Toxicity and therapeutic efficacy of the antibodies of the invention, administered alone or
25 in combination with another therapeutic agent, can be determined by standard pharmaceutical procedures in cell cultures or experimental animals, *e.g.*, for determining the LD₅₀ (the dose lethal to 50% of the population) and the ED₅₀ (the dose therapeutically effective in 50% of the population). The dose ratio between toxic and therapeutic effects is the therapeutic index (LD₅₀/ ED₅₀). The data obtained from these cell culture assays and animal studies can be used in formulating a range of
30 dosage for use in human. The dosage of such compounds lies preferably within a range of circulating concentrations that include the ED₅₀ with little or no toxicity. The dosage may vary within this range depending upon the dosage form employed and the route of administration.

In a further embodiment, a further therapeutic agent that is administered to a subject in association with a multispecific antibody or antigen-binding fragment thereof of the invention in

accordance with the Physicians' Desk Reference 2003 (Thomson Healthcare; 57th edition (November 1, 2002)).

5 The mode of administration can vary. Routes of administration include oral, rectal, transmucosal, intestinal, parenteral; intramuscular, subcutaneous, intradermal, intramedullary, intrathecal, direct intraventricular, intravenous, intraperitoneal, intranasal, intraocular, inhalation, insufflation, topical, cutaneous, transdermal, intra-arterial, epidural or intracranial.

10 In particular embodiments, the multispecific antibodies or antigen-binding fragments thereof of the invention can be administered by an invasive route such as by injection. In further embodiments of the invention, an antibody or antigen-binding fragment thereof, or pharmaceutical composition thereof, is administered intravenously, subcutaneously, intramuscularly, intraarterially, intracranially, epidurally or by inhalation, aerosol delivery. Administration by non-invasive routes (e.g., orally; for example, in a pill, capsule or tablet) is also within the scope of the present invention.

It is preferred to administer the binding molecules of the invention through the intravenous, epidural or intracranial route.

15 The present invention provides a vessel (e.g., a plastic or glass vial, e.g., with a cap or a chromatography column, hollow bore needle or a syringe cylinder) comprising any of the antibodies or antigen-binding fragments of the invention or a pharmaceutical composition thereof. The present invention also provides an injection device comprising any of the antibodies or antigen-binding fragments of the invention or a pharmaceutical composition thereof. An injection device is a device
20 that introduces a substance into the body of a patient via a parenteral route, e.g., intramuscular, subcutaneous, intracranial, epidural or intravenous. For example, an injection device may be a syringe (e.g., pre-filled with the pharmaceutical composition, such as an auto-injector) which, for example, includes a cylinder or barrel for holding fluid to be injected (e.g., antibody or fragment or a pharmaceutical composition thereof), a needle for piercing skin and/or blood vessels for injection of
25 the fluid; and a plunger for pushing the fluid out of the cylinder and through the needle bore. In an embodiment of the invention, an injection device that comprises an antibody or antigen-binding fragment thereof of the present invention or a pharmaceutical composition thereof is an intravenous (IV) injection device. Such a device includes the antibody or fragment or a pharmaceutical composition thereof in a cannula or trocar/needle which may be attached to a tube which may be
30 attached to a bag or reservoir for holding fluid (e.g., saline; or lactated ringer solution comprising NaCl, sodium lactate, KCl, CaCl₂ and optionally including glucose) introduced into the body of the patient through the cannula or trocar/needle. The antibody or fragment or a pharmaceutical composition thereof may, in an embodiment of the invention, be introduced into the device once the trocar and cannula are inserted into the vein of a subject and the trocar is removed from the inserted
35 cannula. The IV device may, for example, be inserted into a peripheral vein (e.g., in the hand or arm); the superior vena cava or inferior vena cava, or within the right atrium of the heart (e.g., a central IV); or into a subclavian, internal jugular, or a femoral vein and, for example, advanced toward the

heart until it reaches the superior vena cava or right atrium (*e.g.*, a central venous line). In an embodiment of the invention, an injection device is an autoinjector; a jet injector or an external infusion pump. A jet injector uses a high-pressure narrow jet of liquid which penetrate the epidermis to introduce the antibody or fragment or a pharmaceutical composition thereof to a patient's body.

5 External infusion pumps are medical devices that deliver the antibody or fragment or a pharmaceutical composition thereof into a patient's body in controlled amounts. External infusion pumps may be powered electrically or mechanically. Different pumps operate in different ways, for example, a syringe pump holds fluid in the reservoir of a syringe, and a moveable piston controls fluid delivery, an elastomeric pump holds fluid in a stretchable balloon reservoir, and pressure from the

10 elastic walls of the balloon drives fluid delivery. In a peristaltic pump, a set of rollers pinches down on a length of flexible tubing, pushing fluid forward. In a multi-channel pump, fluids can be delivered from multiple reservoirs at multiple rates. The present invention also provides sterilization fluids, a sharp bladed instrument, and a drilling device. Sterilization fluids may be used to sterilize the area of the head where the incision is made. A sharp bladed instrument may be a scalpel or another

15 instrument that can be used to make such an incision in the head. The drilling device, may be a drill that can be used to make a small hole into the skull. Through this hole an injection can be made using an injection device as described above.

The pharmaceutical compositions disclosed herein may also be administered with a needleless hypodermic injection device; such as the devices disclosed in U.S. Patent Nos. 6,620,135;

20 6,096,002; 5,399,163; 5,383,851; 5,312,335; 5,064,413; 4,941,880; 4,790,824 or 4,596,556. Such needleless devices comprising the pharmaceutical composition are also part of the present invention. The pharmaceutical compositions disclosed herein may also be administered by infusion. Examples of well-known implants and modules for administering the pharmaceutical compositions include those disclosed in: U.S. Patent No. 4,487,603, which discloses an implantable micro-infusion pump for dispensing medication at a controlled rate; U.S. Patent No. 4,447,233, which discloses a medication

25 infusion pump for delivering medication at a precise infusion rate; U.S. Patent No. 4,447,224, which discloses a variable flow implantable infusion apparatus for continuous drug delivery; U.S. Patent. No. 4,439,196, which discloses an osmotic drug delivery system having multi-chamber compartments. Many other such implants, delivery systems, and modules are well known to those skilled in the art

30 and those comprising the pharmaceutical compositions of the present invention are within the scope of the present invention.

Furthermore, one may administer the antibody or fragment in a targeted drug delivery system, for example, in a liposome coated with a tissue-specific antibody or with an antibody that targets a BBB receptor. The liposomes will be targeted to and taken up selectively by the afflicted

35 tissue. Such methods and liposomes are part of the present invention.

The administration regimen depends on several factors, including the serum or tissue turnover rate of the therapeutic antibody or antigen-binding fragment, the level of symptoms, the immunogenicity of the therapeutic antibody, and the accessibility of the target cells in the biological

matrix. Preferably, the administration regimen delivers sufficient therapeutic antibody or fragment to effect improvement in the target disease state, while simultaneously minimizing undesired side effects. Accordingly, the amount of biologic delivered depends in part on the particular therapeutic antibody and the severity of the condition being treated. Guidance in selecting appropriate doses of therapeutic antibodies or fragments is available (see, e.g., Wawrzynczak (1996) *Antibody Therapy*, Bios Scientific Pub. Ltd, Oxfordshire, UK; Kresina (ed.) (1991) *Monoclonal Antibodies, Cytokines and Arthritis*, Marcel Dekker, New York, NY; Bach (ed.) (1993) *Monoclonal Antibodies and Peptide Therapy in Autoimmune Diseases*, Marcel Dekker, New York, NY; Baert, et al. (2003) *New Engl. J. Med.* 348:601-608; Milgrom et al. (1999) *New Engl. J. Med.* 341:1966-1973; Slamon et al. (2001) *New Engl. J. Med.* 344:783-792; Beniaminovitz et al. (2000) *New Engl. J. Med.* 342:613-619; Ghosh et al. (2003) *New Engl. J. Med.* 348:24-32; Lipsky et al. (2000) *New Engl. J. Med.* 343:1594-1602).

Determination of the appropriate dose is made by the clinician, e.g., using parameters or factors known or suspected in the art to affect treatment. Generally, the dose begins with an amount somewhat less than the optimum dose and it is increased by small increments thereafter until the desired or optimum effect is achieved relative to any negative side effects. Important diagnostic measures include those of symptoms of, e.g., the inflammation or level of inflammatory cytokines produced. In general, it is desirable that a biologic that will be used is derived from the same species as the animal targeted for treatment, thereby minimizing any immune response to the reagent. In the case of human subjects, for example, humanized and fully human antibodies are may be desirable.

Multispecific antibodies or antigen-binding fragments thereof disclosed herein may be provided by continuous infusion, or by doses administered, e.g., daily, 1-7 times per week, weekly, bi-weekly, monthly, bimonthly, quarterly, semiannually, annually etc. Doses may be provided, e.g., intravenously, subcutaneously, topically, orally, nasally, rectally, intramuscular, intracerebrally, intraspinally, or by inhalation. A total weekly dose is generally at least 0.05 µg/kg body weight, more generally at least 0.2 µg/kg, 0.5 µg/kg, 1 µg/kg, 10 µg/kg, 100 µg/kg, 0.25 mg/kg, 1.0 mg/kg, 2.0 mg/kg, 5.0 mg/ml, 10 mg/kg, 25 mg/kg, 50 mg/kg or more (see, e.g., Yang, et al. (2003) *New Engl. J. Med.* 349:427-434; Herold, et al. (2002) *New Engl. J. Med.* 346:1692-1698; Liu, et al. (1999) *J. Neurol. Neurosurg. Psych.* 67:451-456; Portielji, et al. (20003) *Cancer Immunol. Immunother.* 52:151-144). Doses may also be provided to achieve a pre-determined target concentration of the antibody in the subject's serum, such as 0.1, 0.3, 1, 3, 10, 30, 100, 300 µg/ml or more. In other embodiments, a multispecific antibody of the present invention is administered, e.g., subcutaneously or intravenously, on a weekly, biweekly, "every 4 weeks," monthly, bimonthly, or quarterly basis at 10, 20, 50, 80, 100, 200, 500, 1000 or 2500 mg/subject.

As used herein, the term "effective amount" refers to an amount of a multispecific antibody or multispecific antigen-binding fragment thereof of the invention that, when administered alone or in combination with an additional therapeutic agent to a cell, tissue, or subject, is effective to cause a measurable improvement in one or more symptoms of disease. An effective dose further refers to that amount of the antibody or fragment sufficient to result in at least partial amelioration of

symptoms, *e.g.*, memory loss, motor dysfunction or cognitive impairment. When applied to an individual active ingredient administered alone, an effective dose refers to that ingredient alone. When applied to a combination, an effective dose refers to combined amounts of the active ingredients that result in the therapeutic effect, whether administered in combination, serially or simultaneously.

5 An effective amount of a therapeutic will result in an improvement of a diagnostic measure or parameter by at least 10%; usually by at least 20%; preferably at least about 30%; more preferably at least 40%, and most preferably by at least 50%. An effective amount can also result in an improvement in a subjective measure in cases where subjective measures are used to assess disease severity.

10 The present invention also includes a therapeutic method in which a sample of cerebrospinal fluid (CSF) comprising the pathological protein (or a fragment thereof) is obtained from a subject diagnosed with a NDD, is purified and afterwards the purified CSF is reentered into the subject. In this method, the multispecific antibodies and antigen-binding fragments thereof are
15 immobilized on a solid phase such a Sephadex, glass or agarose resin or filter paper, using methods well known in the art. A sample of CSF containing the pathological protein (or a fragment thereof) obtained from a subject diagnosed with an NDD is contacted with the solid phase, hereby the pathological protein is removed and the fluid is re-entered into the subject. Such immobilized antibodies and fragments form part of the present invention.

20 In a further embodiment, nucleic acid constructs are implemented into the cell that encode for the multispecific antibodies of the invention.

For transcription from a nucleic acid construct, a regulatory sequence, such as a promoter, enhancer, terminator, splice donor and acceptor, or polyadenylation site may be used to transcribe the DNA. The RNA strand(s) may or may not be polyadenylated and the RNA strand(s) may or may not be capable of being translated into a polypeptide by the cell's translational apparatus.

25 The nucleic acid construct can be present as plasmid in the nucleus, where it can be translated repeatedly into the multispecific antibodies of the invention. Alternatively, the nucleic acid construct for encoding the multispecific antibodies of the present invention can be stably integrated into the chromosomal DNA of a target cell or progenitor thereof. Stable integration into the host cell DNA is a natural characteristic of various types of viruses, so that genes introduced by these vectors
30 can be maintained for the life of that cell. Furthermore, the vector will be present in all daughter cells that result from cell division. Stable integration may be obtained by using such viruses as retroviruses, lentiviruses, adenoviruses, adeno-associated virus or herpes simplex virus. Suitable retroviruses include murine leukaemia virus (MLV) and moloney murine leukemia virus (Mo-MuLV). The advantages of using a virus such as MLV are that there is no pre-existing immunogenicity to
35 such a virus, that there will be no immune response to viral gene products and that the virions are relatively easy to produce. However, the virus requires dividing cells for infection and may integrate randomly into the host cell genome leading to potential oncogenesis by insertional mutation. Also, it

is known that the LTRs may interfere with gene expression. Suitable lentiviruses for use as the viral vector in a method of the invention include bovine lentiviruses, such as bovine immunodeficiency virus and Jembrana disease virus, equine lentiviruses such as equine infectious anemia virus, feline lentiviruses such as feline immunodeficiency virus (FIV), panther lentivirus and puma lentivirus, 5 ovine/caprine lentiviruses such as Brazilian caprine lentivirus, caprine arthritis-encephalitis virus, caprine lentivirus, Maedi-Visna virus, ovine lentivirus and Visna lentivirus and the primate lentivirus group including such viruses as human immunodeficiency virus (HIV) and simian immunodeficiency virus (SIV). An advantage of the use of lentivirus is that no dividing cells are required and that they may thus be used for *in vivo* applications to transduce non-dividing cells (e.g. 10 memory T-lymphocytes, hemopoietic stem cells, neurons). However, when using viruses that are normally capable of causing infection in humans, these viruses need to be attenuated or made sufficiently harmless by deletion of essential genes.

Use of adeno-associated virus (AAV) vectors in a method of the invention is advantageous because these viruses both expeditiously infect non-dividing cells and may undergo site-specific 15 integration into the host cell genome. More importantly, AAV-delivered DNA may be available as plasmids in the nucleus. Both scenarios result in long-term transduction and may therefore require only a single dose administration. AAV is a non-pathogenic dependent parvovirus with a broad host range, capable of high levels of transduction and expression in the host cell. Moreover, AAVs are particularly useful for delivering genes into the brain. For example, AAV-mediated delivery of genes 20 encoding for anti-tau antibodies and fragments such as scFv into the brain was shown to be an effective tool for reducing tau pathology in mice. Much higher levels of antibody or antibody-fragments were observed in the brain, compared to systemic passive immunization, resulting in a marked decrease in tau accumulation. (Liu, W. et al., J. Neurosci., 2016, 36 (49), 12425-12435, Ising, C. et al, J. Exp. Med., 2017, 214 (5), 1227-1238). The skilled person will know how to administer the 25 virus vector to a subject in need thereof.

Most preferred is the use of viral vectors derived from lentiviruses, adeno-associated virus (AAV), or combinations such as adenovirus-AAV hybrid vectors. The methods for constructing viral vectors and packaging those into a viral particle are known to a person skilled in the art. See Ising, C. et al, J. Exp. Med., 2017, 214 (12), 1; Stadler, C.R., Nature Medicine, 2017, 23, 815; 30 WO2016126993 A1.

Any regulatory sequences which are known or are found to cause expression of the therapeutic gene in the target cell can be used in the present invention. Such regulatory sequences may for instance be obtained from humans, animals, plants or fungi, or their associated viruses, or may be chemically synthesized, but are preferably target cell specific and include suitable eukaryotic 35 or viral promoters operably linked to the therapeutic gene and active in directing its transcription in the target cells as well as terminators. The promoters include, but are not limited to, promoters from organ- or tissue-specific genes, or promoters of constitutively expressed genes. Examples of suitable promoters for directing the transcription of the therapeutic gene in mammalian cells are the SV40

promoter (Subramani et al., Mol. Cell Biol. 1 (1981), 854-864), the MT-1 (metallothionein gene) promoter (Palmiter et al., Science 222 (1983), 809-814) or the adenovirus 2 major late promoter the 5'-long terminal repeats from retroviruses and lentiviruses, the cytomegalus virus (CMV) immediate early promoter, and the like. These promoters and various others are easily obtainable for a person skilled in the art. Other regulatory sequences include terminator sequences and polyadenylation signals, including every sequence functioning as such in the target cells.

Kits

Further provided are kits comprising one or more components that include, but are not limited to, a multispecific antibody or antigen-binding fragment, as discussed herein in association with one or more additional components including, but not limited to a pharmaceutically acceptable carrier and/or a therapeutic agent, as discussed herein. The antibody or fragment and/or the therapeutic agent can be formulated as a pure composition or in combination with a pharmaceutically acceptable carrier, in a pharmaceutical composition.

In one embodiment, the kit includes a multispecific antibody or antigen-binding fragment thereof of the invention or a pharmaceutical composition thereof in one container (e.g., in a sterile glass or plastic vial) and a pharmaceutical composition thereof and/or a therapeutic agent in another container (e.g., in a sterile glass or plastic vial).

In another embodiment, the kit comprises a combination of the invention, including a multispecific antibody or antigen-binding fragment thereof of the invention along with a pharmaceutically acceptable carrier, optionally in combination with one or more therapeutic agents formulated together, optionally, in a pharmaceutical composition, in a single, common container.

If the kit includes a pharmaceutical composition for parenteral administration to a subject, the kit can include a device for performing such administration. For example, the kit can include one or more hypodermic needles or other injection devices as discussed above.

The kit can include a package insert including information concerning the pharmaceutical compositions and dosage forms in the kit. Generally, such information aids patients and physicians in using the enclosed pharmaceutical compositions and dosage forms effectively and safely. For example, the following information regarding a combination of the invention may be supplied in the insert: pharmacokinetics, pharmacodynamics, clinical studies, efficacy parameters, indications and usage, contraindications, warnings, precautions, adverse reactions, overdosage, proper dosage and administration, how supplied, proper storage conditions, references, manufacturer/distributor information and patent information.

Detection Kits and Therapeutic Kits

As a matter of convenience, a multispecific antibody or antigen-binding fragment thereof of the invention can be provided in a kit, i.e., a packaged combination of reagents in predetermined amounts with instructions for performing the diagnostic or detection assay. Where the antibody or
5 fragment is labeled with an enzyme, the kit will include substrates and cofactors required by the enzyme (*e.g.*, a substrate precursor which provides the detectable chromophore or fluorophore). In addition, other additives may be included such as stabilizers, buffers (*e.g.*, a block buffer or lysis buffer) and the like. The relative amounts of the various reagents may be varied widely to provide for concentrations in solution of the reagents which substantially optimize the sensitivity of the assay.
10 Particularly, the reagents may be provided as dry powders, usually lyophilized, including excipients which on dissolution will provide a reagent solution having the appropriate concentration.

Also provided are diagnostic or detection reagents and kits comprising one or more such reagents for use in a variety of detection assays, including for example, immunoassays such as ELISA (sandwich-type or competitive format). The kit's components may be pre-attached to a solid support,
15 or may be applied to the surface of a solid support when the kit is used. In some embodiments of the invention, the signal generating means may come pre-associated with an antibody or fragment of the invention or may require combination with one or more components, *e.g.*, buffers, antibody-enzyme conjugates, enzyme substrates, or the like, prior to use. Kits may also include additional reagents, *e.g.*, blocking reagents for reducing nonspecific binding to the solid phase surface, washing reagents,
20 enzyme substrates, and the like. The solid phase surface may be in the form of a tube, a bead, a microtiter plate, a microsphere, or other materials suitable for immobilizing proteins, peptides, or polypeptides. In particular aspects, an enzyme that catalyzes the formation of a chemiluminescent or chromogenic product or the reduction of a chemiluminescent or chromogenic substrate is a component of the signal generating means. Such enzymes are well known in the art. Kits may comprise any of
25 the capture agents and detection reagents described herein. Optionally the kit may also comprise instructions for carrying out the methods of the invention.

Also provided is a kit comprising a multispecific antibody or antigen-binding fragment thereof described herein packaged in a container, such as a vial or bottle, and further comprising a label attached to or packaged with the container, the label describing the contents of the container
30 and providing indications and/or instructions regarding use of the contents of the container to treat one or more disease states as described herein.

In one aspect, the kit is for treating NDDs and comprises a multispecific antibody or antigen-binding fragment thereof and a further therapeutic agent or a vaccine. The kit may optionally further include a syringe for parenteral, *e.g.*, intravenous, administration. In another aspect, the kit
35 comprises a multispecific antibody or antigen-binding fragment thereof and a label attached to or packaged with the container describing use of the antibody or fragment with the vaccine or further therapeutic agent. In yet another aspect, the kit comprises the vaccine or further therapeutic agent

and a label attached to or packaged with the container describing use of the vaccine or further therapeutic agent with the multispecific antibody or fragment. In certain embodiments, the multispecific antibody and vaccine or further therapeutic agent are in separate vials or are combined together in the same pharmaceutical composition.

5 The therapeutic and detection kits disclosed herein may also comprise at least one of the antibody, peptide, antigen-binding fragment, or polynucleotide disclosed herein and instructions for using the composition as a detection reagent or therapeutic agent. Containers for use in such kits may typically comprise at least one vial, test tube, flask, bottle, syringe or other suitable container, into which one or more of the detection and/or therapeutic composition(s) may be placed, and preferably
10 suitably aliquoted. Where a second therapeutic agent is also provided, the kit may also contain a second distinct container into which this second detection and/or therapeutic composition may be placed. Alternatively, a plurality of compounds may be prepared in a single pharmaceutical composition, and may be packaged in a single container means, such as a vial, flask, syringe, bottle, or other suitable single container. The kits disclosed herein will also typically include a means for
15 containing the vial(s) in close confinement for commercial sale, such as, *e.g.*, injection or blow-molded plastic containers into which the desired vial(s) are retained. Where a radiolabel, chromogenic, fluorogenic, or other type of detectable label or detecting means is included within the kit, the labeling agent may be provided either in the same container as the detection or therapeutic composition itself, or may alternatively be placed in a second distinct container means into which this second
20 composition may be placed and suitably aliquoted. Alternatively, the detection reagent and the label may be prepared in a single container means, and in most cases, the kit will also typically include a means for containing the vial(s) in close confinement for commercial sale and/or convenient packaging and delivery.

 A device or apparatus for carrying out the detection or monitoring methods described
25 herein is also provided. Such an apparatus may include a chamber or tube into which sample can be input, a fluid handling system optionally including valves or pumps to direct flow of the sample through the device, optionally filters to separate plasma or serum from blood, mixing chambers for the addition of capture agents or detection reagents, and optionally a detection device for detecting
30 the amount of detectable label bound to the capture agent immunocomplex. The flow of sample may be passive (*e.g.*, by capillary, hydrostatic, or other forces that do not require further manipulation of the device once sample is applied) or active (*e.g.*, by application of force generated via mechanical pumps, electroosmotic pumps, centrifugal force, or increased air pressure), or by a combination of active and passive forces.

 In further embodiments, also provided is a processor, a computer readable memory, and
35 a routine stored on the computer readable memory and adapted to be executed on the processor to perform any of the methods described herein. Examples of suitable computing systems, environments, and/or configurations include personal computers, server computers, hand-held or laptop devices, multiprocessor systems, microprocessor-based systems, set top boxes, programmable consumer

electronics, network PCs, minicomputers, mainframe computers, distributed computing environments that include any of the above systems or devices, or any other systems known in the art.

Example 1: Production of bispecific antibodies

5 Production of bispecific antibodies was performed at Absolute Antibody (Oxford, UK). The produced bispecific antibodies consist of a mouse IgG1 molecule with two scFv molecules fused at the C-terminus of the heavy chain. Variable domains from publicly available DNA sequences (see Table 1) were designed and optimised for expression in mammalian cells (HEK293) prior to being synthesized. The sequences were then subcloned into Absolute Antibody cloning and expression vectors for mouse IgG1 and scFv. HEK293 cells were passaged to the optimum stage for transient transfection. Cells
10 were transiently transfected with expression vectors and cultured for a further 6-14 days. An appropriate volume of cells was transfected with the aim of obtaining 1-5 mg of purified antibody. Cultures were harvested and a one-step purification was performed using affinity chromatography. The antibodies were analyzed for purity by SDS-PAGE and the concentration was determined by UV spectroscopy. This format of the resulting bispecific antibodies is commonly referred to as IgG-scFv,
15 IgG-scFv (HC) or BiS3. The bispecific antibodies were obtained (Table 2) and characterized using SDS page under both reducing and non-reducing conditions (Figure 1). Under non-reducing conditions the bispecific antibodies have the expected molecular weight of approximately 199kDa. Under reducing conditions IgG fragments were observed at expected molecular weights.

Table 1; CDR's used for preparing bispecific antibodies

clone	CDR's	publication
Hu-37D3-H9.v28	SYGMS (CDR-1 (HC)) TINSGGTYTYYPDSVKG (CDR-2 (HC)) SYSGAMDY (CDR-3 (HC)) RSSQSIVHSNGNTYFE (CDR-1 (LC)) KVSNRFS (CDR-2 (LC)) FQGS LVPWT (CDR-3 (LC)) EVQLVESGGG LVQPGGSLRL SCAASGLIFR SYGMSWVRQAPGKGLEWVAT INSGGTYTY PDSVKGRFTI SRDNSKNTLYLQMNSLRAED TAVYYCANSY SGAMDYWGQG TLVTVSS (HC) DDVLTQTPLS LPVTPGQPAS ISCRSSQSIV HSNGNTYLEWYLQKPGQSPQ LLIYKVSNR SGVPDRFSGS GSGTDFTLKISRVEAEDVGV YYCFQGS LVP WTPGQGTKVE IK (LC)	WO2016196726
AB1	EVQLQESGPGLVKPSETLSLTCTVSGFSLTSNDI AWIRQPPGKGLEWMGTIWDGSTNYNAAVQS RVTISVDTSKNQFSLKLSSVTAADTAVYYCAR HRLYYGAFDYWGQGTMTVTVSS (HC) DIVMSQSPSSLAVSVGEKVTMSCKSIQTLLYSS NQKNYLAWFQQKPGQSPKLLIYWASIRKSGVP DRFTGSGSGTDFLTISVKAEDLAVYYCQY YSYPLTFGAGTKLELK (LC)	WO2017/005734

m9E4	NYGMS (CDR-1 (HC)) SISSGGGSTYYPDNVKG (CDR-2 (HC)) GGAGIDY (CDR-3 (HC)) KSIQTLTYSSNQKNYLA (CDR-1 (LC)) WASIRKS (CDR-2 (LC)) QQYYSYPLT (CDR-3 (LC)) EVKLVESGGGLVKPGASLKLSCAASGFTFSNY GMSWVRQTSDKRLEWVASISSGGGSTYYPDN VKGRFTISREDAKNTLYLQMSSLRSEDTALYY CSRGGAGIDYWGQGTTLTVSS (HC) DIVMSQSPSSLAVSVGEKVTMSCKSIQTLLYSS NQKNYLAWFQQKPGQSPKLLIYWASIRKSGVP DRFTGSGSGTDFLTISSVKAEDLAVYYCQQY YSYPLTPGAGTKLELK (LC)	US 20130108546 A1
M1	GYHFTSYWMH (CDR-1 (HC)) VIHPNSGSINYNEKFES (CDR-2 (HC)) ERDSTEVLPM DY (CDR-3 (HC)) RASKSINKYLA (CDR-1 (LC)) SGSTLQS (CDR-2 (LC)) QQHNEYPLT (CDR-3 (LC)) QVQLQQPGAELVKPGASVKLSCKSSGYHFTSY WMHWVKQRPGQGLEWIGVIHPNSGSINYNEK FESKATLTVDKSSSTAYMQLSSLTSEDSAVYYC AGERDSTEVLPM DYWGQGTSTVTVSS (HC) DVQITQSPSYLAASPGETITINCRASKSINKYLA WYQKPGKTNKLLIYSGSTLQSGIPSRFSGSGS GTDFTLTISLEPEDFAMYQCQHNEYPLTFGA GTKLELK (LC)	US20160355574A1

Table 2; bispecific antibodies

ID	Clone 1 (IgG)	Antigen 1	Clone 2 (scFv)	Antigen 2	Purified (mg)	Yield (mg/L)
bAb0117	m9E4	Alpha-synuclein	hu37D3-H9.v28	Tau	5.7	14
bAb0119	AB1	Tau	M1	C1q	5.2	13
bAb0120	M1	C1q	AB1	Tau	16.6	41

Example 2; Evaluation of binding of the bispecific antibodies to protein targets

The bispecific antibodies obtained with Example 1 were evaluated in an ELISA assay in terms of binding to human tau, human alpha-synuclein or human C1q.

The ELISA plates were coated with with recombinant human tau (R&D Systems, SP-495), human α -synuclein (R&D Systems, SP-485-500) or human C1q (Sigma, C1740) at 5mg/ml. Serial dilutions of 3-fold were made for each bispecific antibody. Negative controls contained no primary antibody. Signal was detected with anti-mouse IgG HRP. The results are shown in Figure 2-4. It is shown that bAb0117 binds both alpha-synuclein and tau, bAb0119 binds both tau and C1q and bAb0120 binds both C1q and Tau.

Example 3; Histological detection of tau and α -synuclein in the brain of transgenic animals.

Tau and alpha-synuclein are intrinsically disordered proteins with complex and natively unfolded structures in vivo. Bispecific antibodies were therefore tested for binding in the brains of transgenic mice that model tau pathology (TMHT, QPS Austria) and alpha-synuclein pathology (Line 61, QPS Austria). These mouse models overexpress a form of human tau and human alpha-synuclein, respectively. Both mouse models accumulate sarkosyl insoluble phosphorylated protein aggregates in neurons, which resemble human tauopathies or synucleinopathies. Stainings were compared to non-transgenic mice. All mice were 12 months of age. Sections of 35 μ m thickness were stained with bispecific antibody (5 μ g/ml). Pathological accumulation of tau was stained on the same section as the bispecific antibody with rabbit anti-tau p214. Pathological accumulation of alpha-synuclein was stained on the same section as the bispecific antibody with rabbit anti-alpha-synuclein p129. Signal was detected with anti-mouse IgG 488 and anti-rabbit IgG Alexa 546. Transgenic and wild-type brain slices were imaged with the same laser power and gain on a Zeiss LSM 700 confocal microscope. The results are shown in Figures 5-8.

The immunofluorescent stainings show that bAb0117 detects accumulated alpha-synuclein in the cortex of 12 months old Line 61 mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated alpha-synuclein that is phosphorylated at residue 129 - which is a marker for alpha-synuclein pathology and not present in nontransgenic animals. Furthermore,

5 bAb0117 detects accumulated tau in the hippocampus of 12 months old TMHT mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated tau that is phosphorylated at residue 214 - which is a marker for tau pathology and not present in nontransgenic animals.

Further, immunofluorescent staining shows that bAb0119 detects accumulated tau in the

10 hippocampus of 12 months old TMHT mice, but not in age-matched controls. The same neurons display immunoreactivity for accumulated tau that is phosphorylated at residue 214 - which is a marker for tau pathology and not present in nontransgenic animals.

Moreover, immunofluorescent staining shows that bAb0120 detects accumulated tau in the hippocampus of 12 months old TMHT mice, but not in age-matched controls. The same neurons

15 display immunoreactivity for accumulated tau that is phosphorylated at residue 214 - which is a marker for tau pathology.

Example 4; Thioflavin T tau aggregation assay

To demonstrate that bispecific antibodies retain their functional properties, they were compared to

20 their monospecific parental antibodies in a tau aggregation assay. Recombinant Tau441 (2N4R) P301L (Analytik Jena, T-1014-1) at 1 μ M final concentration, was incubated with 30 μ M sodium octadecylsulfate (ODS) and 1 μ M Heparin in reagent buffer (20 μ M Thioflavine T, 5 mM 1,4-dithioerythritol (DTT), 100 mM sodium chloride, 10 mM HEPES pH 7.4) for 15 hrs at 37°C in black no-binding 96 well plates. IgG and IgG-scFv were used in the same concentration as recombinant tau

25 (1 μ M). The scFv was used at 2 μ M, because IgG-scFv molecules contain 2 scFv portions. Compounds were incubated with the before mentioned tau-

Heparin-ODS-Buffer solution. Six technical replicates were performed. Immediately after preparation a baseline measurement was carried out and following 4 and 15 hours of incubation at 37°C, fluorescence was again detected by using 450 nm excitation and 485 nm emission. The results are

30 shown in Figure 9. No tau aggregation was observed when no tau was added (lane 1; no Tau), but was observed in the presence of tau without antibodies (lane 10; VC). No inhibition of tau aggregation was observed with negative control antibody 2B11 (rPeptide) that binds to tau phosphorylated at residue 231 - which is absent on recombinant tau used in this assay (lane 8; 2B11). Positive control 3E8-1A6 (Wako) binds to the repeat domain of tau - which is necessary for aggregation - and completely

35 inhibits tau aggregation (lane 9; 3E8-1A6). Both the monospecific (lane 2; hu37D3-H9.v28) and bispecific antibody (lane 3; M9E4/hu37D3-H9.v28) that bind to the distal N-terminal of tau show modest inhibition of aggregation. The bispecific variant of this antibody shows slightly stronger inhibition of tau aggregation. Antibody AB1 shows complete inhibition of tau aggregation as IgG (lane 4; AB1), but not as scFv (lane 5; AB1). The bispecific variants of AB1 showed complete inhibition of

tau aggregation when AB1 was used as IgG (lane 6; AB1/M1) or added as scFv to M1 IgG (lane 7; M1/Ab1).

Claims

1. A multispecific binding molecule, preferably an antibody, comprising at least a first binding site binding to a first target selected from the group consisting essentially of human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, TDP43, mHTT, and fragments thereof, and at least a second binding site binding to a second target selected from the group consisting essentially of α -synuclein, TDP43, mHTT, human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, and fragments thereof, wherein the first and second target are different.
2. A multispecific binding molecule according to claim 1, wherein said binding molecule comprises at least a first binding site binding to human tau protein or post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof and at least a second binding site binding to α -synuclein or a fragment thereof.
3. A multispecific binding molecule according to claim 2, wherein said binding molecule comprises at least a first binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or V_L domains and/or V_H domains of BIIB092, C2N 8E12, RG7345, 37D3-H9, 54C1-H11, 123E9, 24A11, Hu-37D3, Hu-125B11, Hu-94B2, DC8E8, Tau13, HT7, BIIB076, 5A6 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%, even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains and a second binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or V_L domains and/or V_H domains of PRX002/9E4, BIIB054, NI-202.3G12, NI-202.12F4, NI-202.3D8, GM285, GM37, BA1, BA2, BA3, BA4, 5C1, 9E4, Syn-211, H3C or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%, even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains.
4. A multispecific binding molecule according to claim 2 or 3, wherein the first binding site is the combinations of CDR sequences and/or V_L domains and/or V_H domains of hu37D3-H9.v28 and the second binding site is the combinations of CDR sequences and/or V_L domains and/or V_H domains of M9E4.

5. A multispecific binding molecule according to claim 4, wherein the multispecific binding molecule has a format of IgG-scFv, preferably wherein the combinations of CDR sequences and/or V_L domains and/or V_H domains of hu37D3-H9.v28 is scFv.
6. A multispecific binding molecule according to claim 2, wherein said binding molecule
 5 comprises at least one binding site binding to human tau protein selected from the group comprising binding fragments of tau13 and binding fragments of 5A6 and at least one binding site binding to α -synuclein selected from the group comprising binding fragments of Syn211 and binding fragments of H3C
7. A multispecific binding molecule according to claim 1, wherein said at least a second
 10 binding site binds to a second target selected from the group C1q, C5a, IL1B, IL6, TNF- α , APOE, IL12 and IL23.
8. A multispecific binding molecule according to claim 7, wherein said binding molecule comprises at least a first binding site binding to human tau protein or post-translationally modified human tau protein, such as phosphorylated, acetylated, glycosylated, glycated, prolyl-isomerized,
 15 nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof and at least a second binding site binding to C1q or a fragment thereof.
9. A multispecific binding molecule according to claim 7 or 8, wherein said binding molecule comprises at least a first binding site comprising a binding sequence selected from the group
 20 consisting of the combinations of CDR sequences and/or the V_L domains and/or V_H domains of BIIB092, C2N 8E12, RG7345, 37D3-H9, 54C1-H11, 123E9, 24A11, Hu-37D3, Hu-125B11, Hu-94B2, DC8E8, Tau13, HT7, BIIB076, 5A6 or fragments thereof having at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at least 96%, even more preferably at least 97%, even more preferably at least
 25 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains and a second binding site comprising a binding sequence selected from the group consisting of the combinations of CDR sequences and/or the V_L domains and/or V_H domains of ANX005 (ANX-M1) and JL-1 or fragments thereof at least 75%, preferably at least 80%, more preferably at least 85%, even more preferably at least 90%, even more preferably at least 95%, even more preferably at
 30 least 96%, even more preferably at least 97%, even more preferably at least 98% or even more preferably at least 99% sequence identity to any one of said V_L domains or V_H domains.
10. A multispecific binding molecule according to claim 8 or 9, wherein said binding molecule comprises at least one binding site binding to tau protein selected from the group comprising binding fragments of tau13 and binding fragments of 5A6 and at least one binding site binding to C1q
 35 selected from the group comprising binding fragments of JL-1 and binding fragments produced by the hybridomas 23B6C8, 5B5C22, 12A5B7 and 4A4B11.
11. A multispecific binding molecule according to claim 8, wherein the first binding site is Ab1 and the second binding site is ANX-M1.
12. A multispecific binding molecule according to claim 11, wherein the multispecific
 40 binding molecule has a format of IgG-scFv.

13. A multispecific binding molecule according to claim 7, wherein said binding molecule comprises at least a first binding site binding to α -synuclein or a fragment thereof and at least a second binding site binding to C1q or a fragment thereof.
14. A multispecific binding molecule according to claim 13, wherein said binding molecule
5 comprises at least one binding site binding to C1q selected from the group comprising binding fragments of JL-1 and binding fragments produced by the hybridomas 23B6C8, 5B5C22, 12A5B7 and 4A4B11 and at least one binding site binding to α -synuclein selected from the group comprising binding fragments of Syn211 and binding fragments of H3C.
15. A multispecific binding molecule according to claims 1-14, wherein said binding
10 molecule comprises a moiety that enables shuttling of the binding molecule through the blood brain barrier, preferably wherein said moiety is a binding site.
16. A multispecific binding molecule according to claims 1-15, wherein said binding molecule is humanized.
17. A multispecific binding molecule according to claims 1-16, wherein said binding
15 molecule is bispecific.
18. A multispecific binding molecule according to claims 1-16, wherein said binding molecule is trispecific.
19. A multispecific binding molecule according to claims 1-18, having a format selected from the group consisting of multispecific binding formats listed in Figure 2 of Brinkmann, et al., MAbs, 2017, 9:182-212 and Figure 1 of Spiess, et al.; Molecular Immunology, 2015, 67:95-106, and multispecific antibody conjugates, for example dual-variable-domain (DVD) antibody, trispecific IgG2 and tetraspecific IgG2, triple-targeting triplebody, triabody, tribody, trispecific triple heads, trispecific triple dAb, tetraspecific dAb, multispecific dAb, circular dimeric single-chain diabody (CD-scDb), linear dimeric single-chain diabody (LD-scDb), disulfide-stabilized Fv fragment, bis-scFv, tandem tri-
25 scFv, bispecific Fab2, Fab3, chemical conjugate trimeric Fab, di-miniantibody, tetraabody, IgG-scFab, scFab-dsscFv, Fv2-Fc, IgG-scFv fusions, such as Bs1Ab, Bs2Ab, Bs3Ab, Trispecific C-terminal fusion, Tri-specific N-terminal fusion, Ts1Ab, Ts2Ab, IgA1, IgA2, IgD, IgE, IgG1, IgG2, IgG3, IgG4, IgM, CODV-Ig, sdAb, trispecific Zybody, tetraspecific Zybody, pentaspecific Zybody, sextaspecific Zybody, septaspecific Zybody, octaspecific Zybody, Knob-into-holes molecules and duobodies.
20. A multispecific binding molecule according to claims 1-18, wherein the multispecific binding molecule has the format selected from the group comprising trispecific triabodies, trispecific tribodies, IgG(H)-scFv, scFv-(H)IgG, IgG(L)-scFv, scFv-(L)IgG, 2scFv-IgG, IgG-2scFv, DVI-IgG, scFab-Fc(kih)-scFv2, scFab-Fc(kih)-scFv, IgG-taFv, scFv4-IgG and TVD-Ig.
21. A multispecific binding molecule according claims 1-20, wherein the multispecific
35 binding molecule is an antibody and wherein the Fc region of the antibody comprises a mutation at one or more of the following positions: 233, 234, 235, 236, 237, 268, 269, 270, 254, 254, 294, 297, 298, 300, 318, 320, 322, 327, 329, 331, as determined with the Kabat numbering scheme.
22. A multispecific binding molecule according to claims 1-21 for use in treatment or prevention of neurodegenerative disorders, selected from the group comprising Alzheimer's Disease,
40 Lewy Body Dementia, Parkinson's Disease, Huntington Disease, Amyotrophic Lateral Sclerosis

(ALS), Frontotemporal Dementia, Frontotemporal Dementia with Parkinsonism-17, Multiple System Atrophy, Corticobasal Degeneration, Progressive Supranuclear Palsy, Pick's Disease, Primary Age Related Tauopathy, Argyrophilic Grain Disease and Cerebral Amyloid Angiopathy.

23. A multispecific binding molecule according to claims 1-21 for use in treatment of
- 5 Alzheimer's Disease.
24. Binding molecules for use according to claim 23, wherein said multispecific binding molecule is a bispecific binding molecule.
25. Binding molecules for use according to claim 23, wherein said multispecific binding molecule is a trispecific binding molecule.
- 10 26. Binding molecules for use according to claim 23, wherein said multispecific binding molecule is a tetraspecific binding molecule.
27. Use of a therapeutic delivery vehicle, encoding any of the multispecific binding molecules according to claims 1-21 in therapy of neurodegenerative disorders.
28. A method for the treatment of neurodegenerative disorders, comprising: administration
- 15 of a multispecific binding molecule according to claims 1-21 to a subject in need thereof.
29. Use according to claim 27, wherein the therapeutic delivery vehicle is an adeno-associated virus vector.
30. A method for treatment of the neurodegenerative disorders, comprising: administration of a nucleic acid construct encoding a multispecific binding molecule according to claims 1-21 in a
- 20 therapeutic delivery vehicle to a subject in need thereof.
31. A diagnostic method for the detection of neurodegenerative disorders, comprising: adding a multispecific binding molecule according to claims 1-21 to a sample obtained from a subject; determining binding of said binding molecule to any of the targets selected from human tau protein and post-translationally modified human tau protein, such as phosphorylated, acetylated,
- 25 glycosylated, glycated, prolyl-isomerized, nitrated, polyaminated, ubiquitinated, sumoylated, oxidated and aggregated human tau protein and cleaved and truncated versions thereof, α -synuclein, , TDP43, mHTT, C1q, C5a, IL1B, IL6, TNF- α , APOE, TREM2, IL12, IL23 and fragments thereof in said sample; diagnosis of neurodegenerative disease if said binding is detected.
32. A kit for the diagnosis of neurodegenerative disorders, comprising a multispecific
- 30 binding molecule according to any of claims 1-21 and means for detection of said binding molecule.

Fig. 1

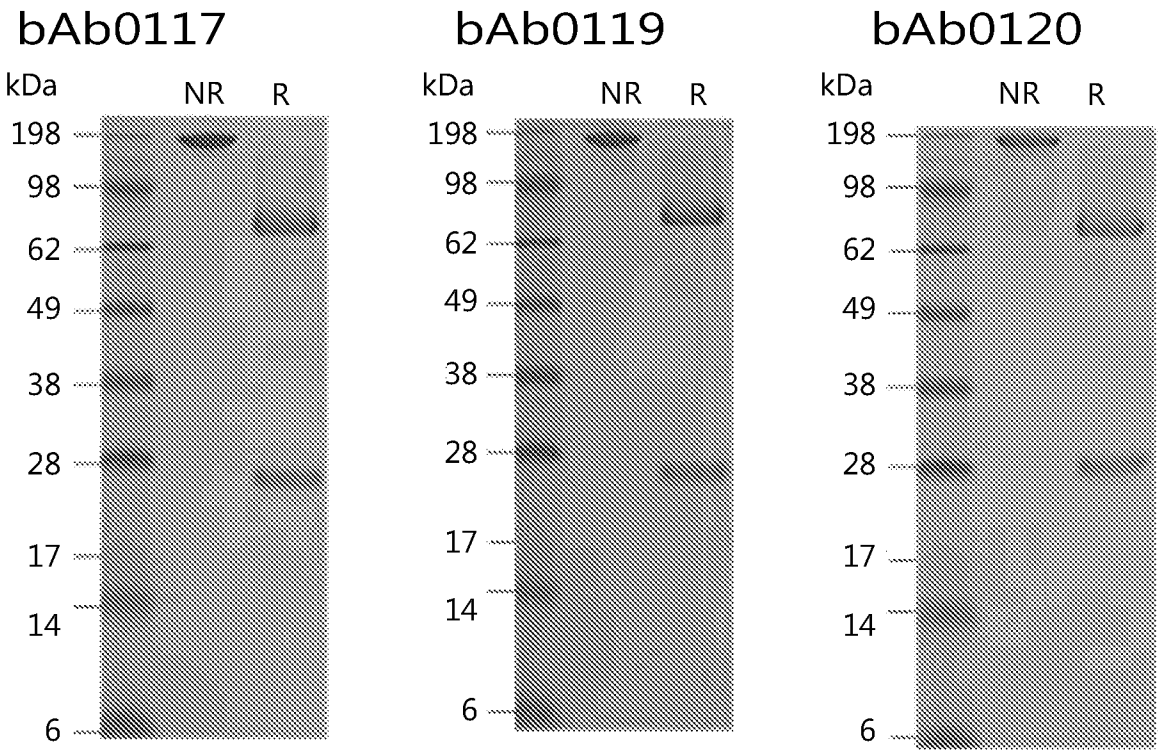


Fig. 2

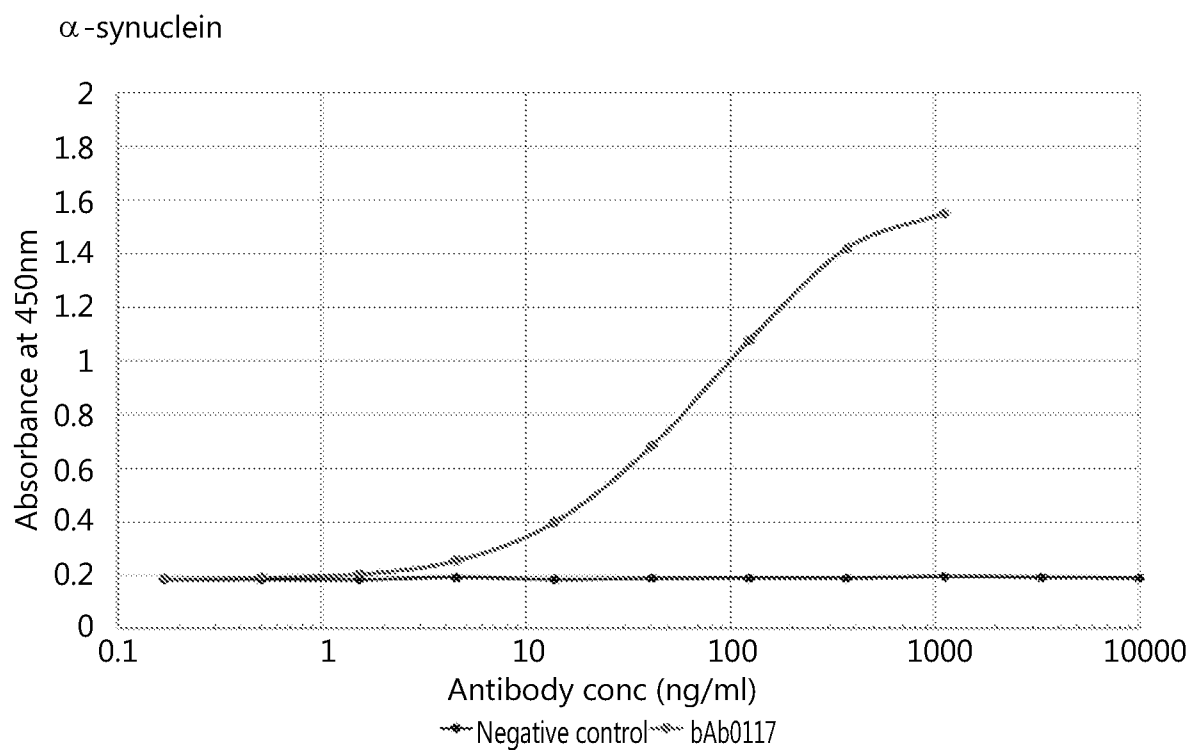
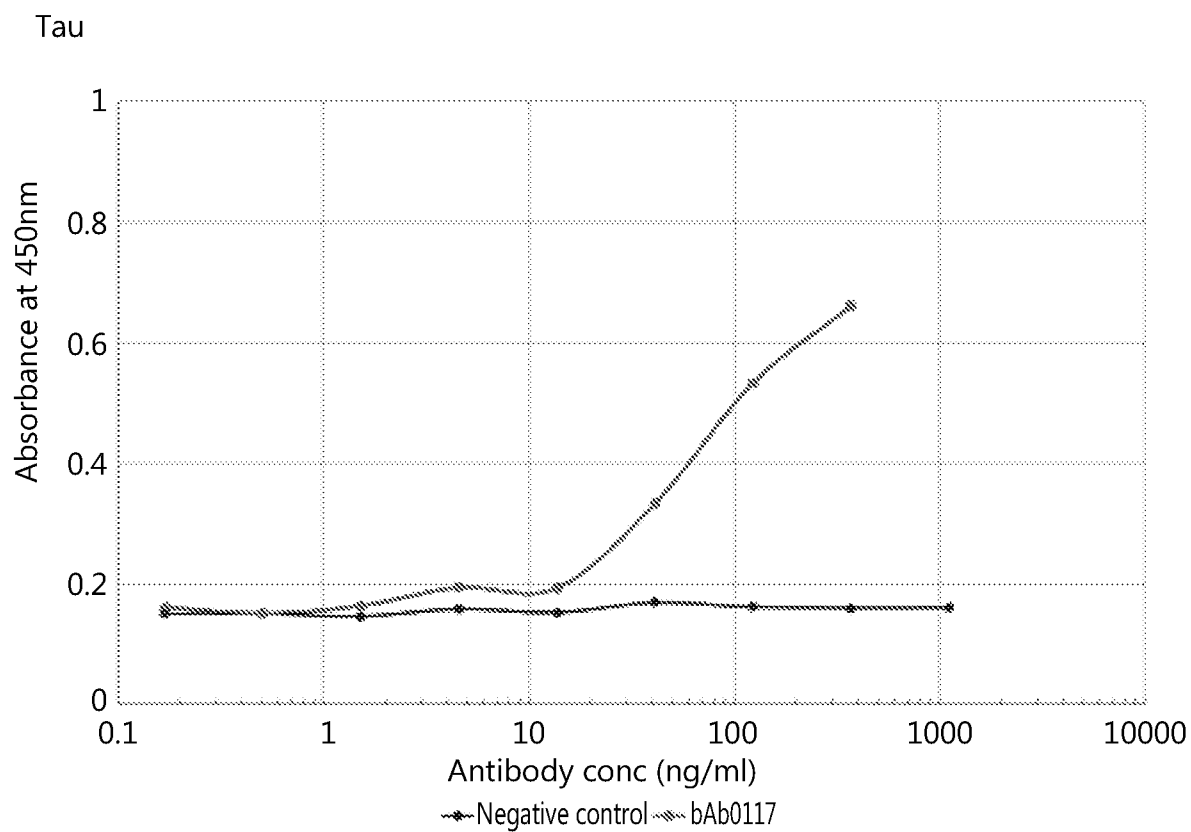
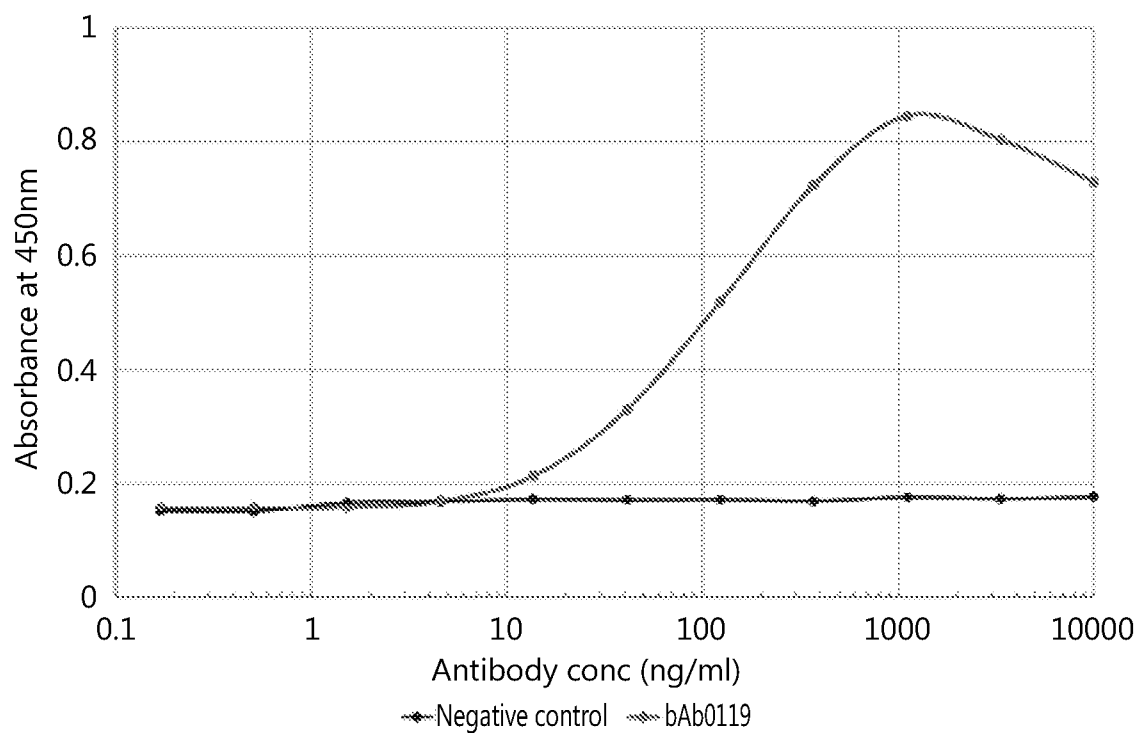
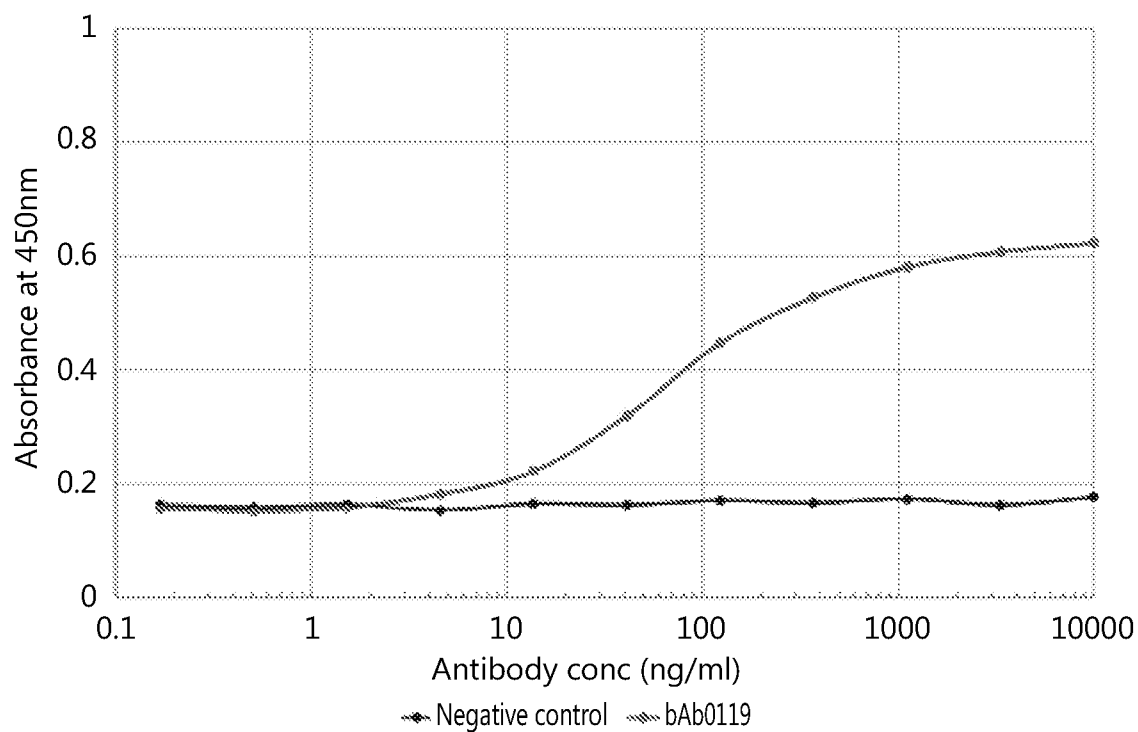


Fig. 3

Tau



C1q



4/7

Figure 4

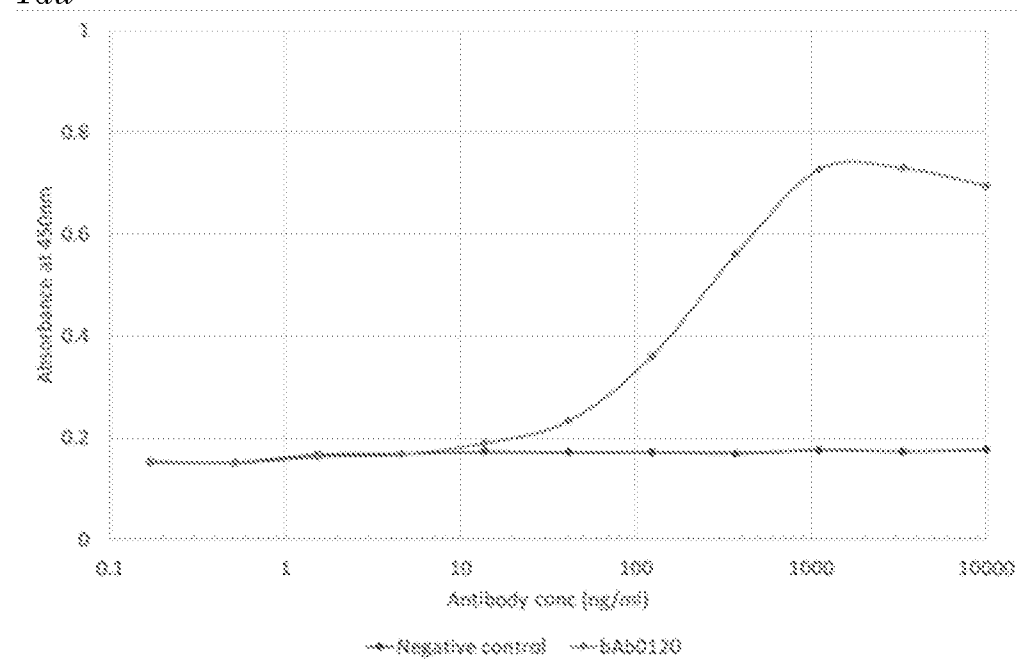
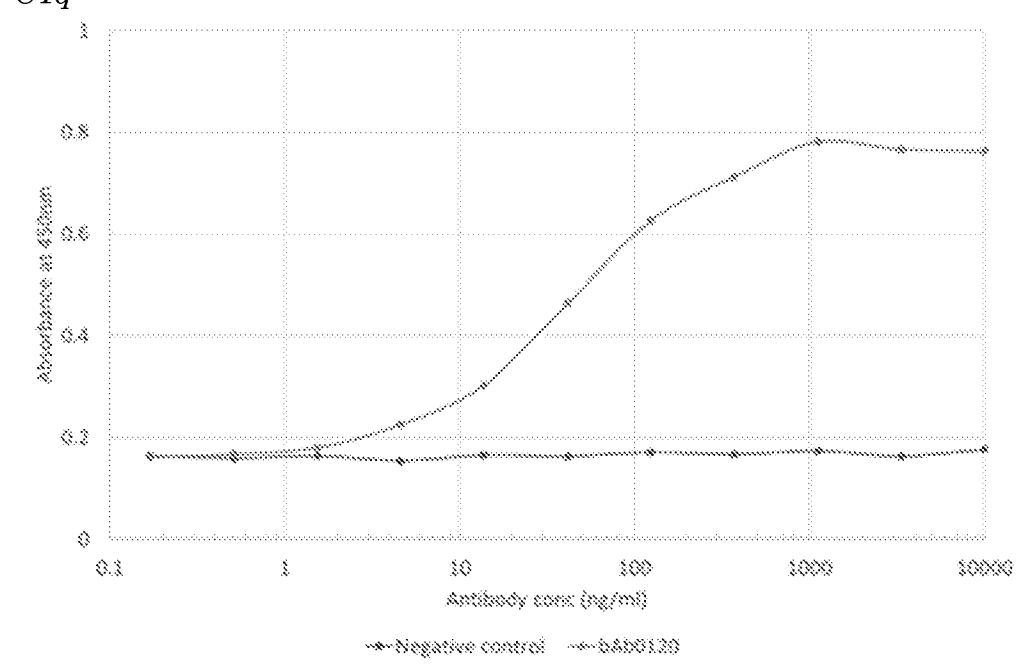
Tau*C1q*

Figure 5

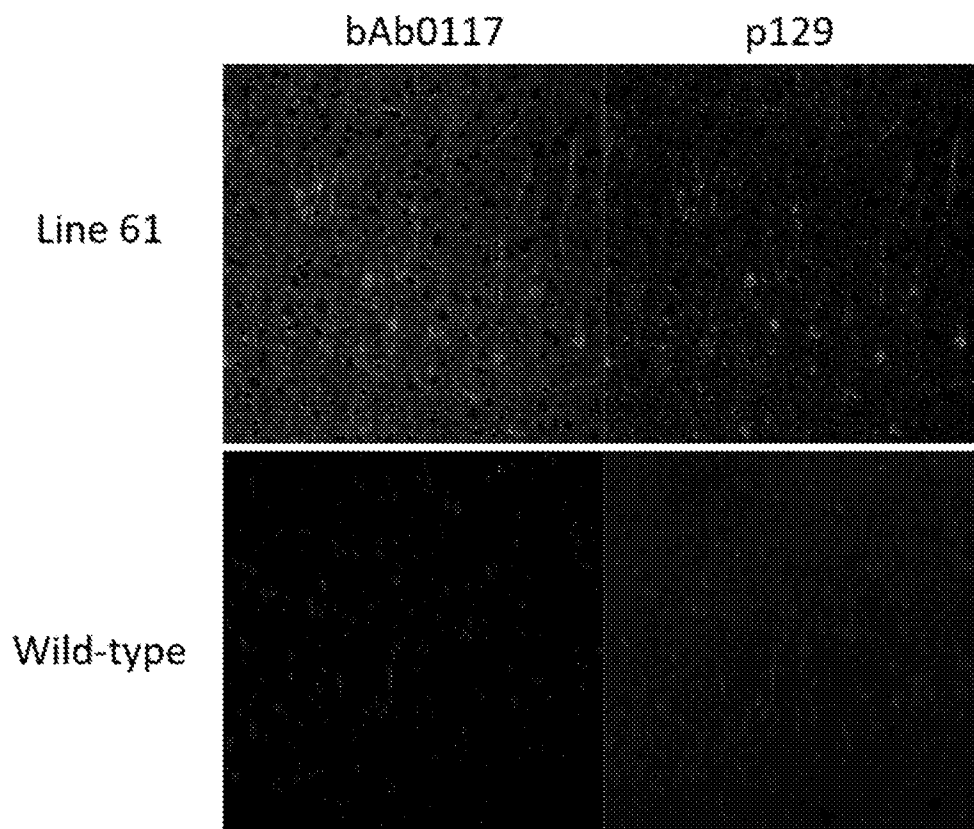


Figure 6

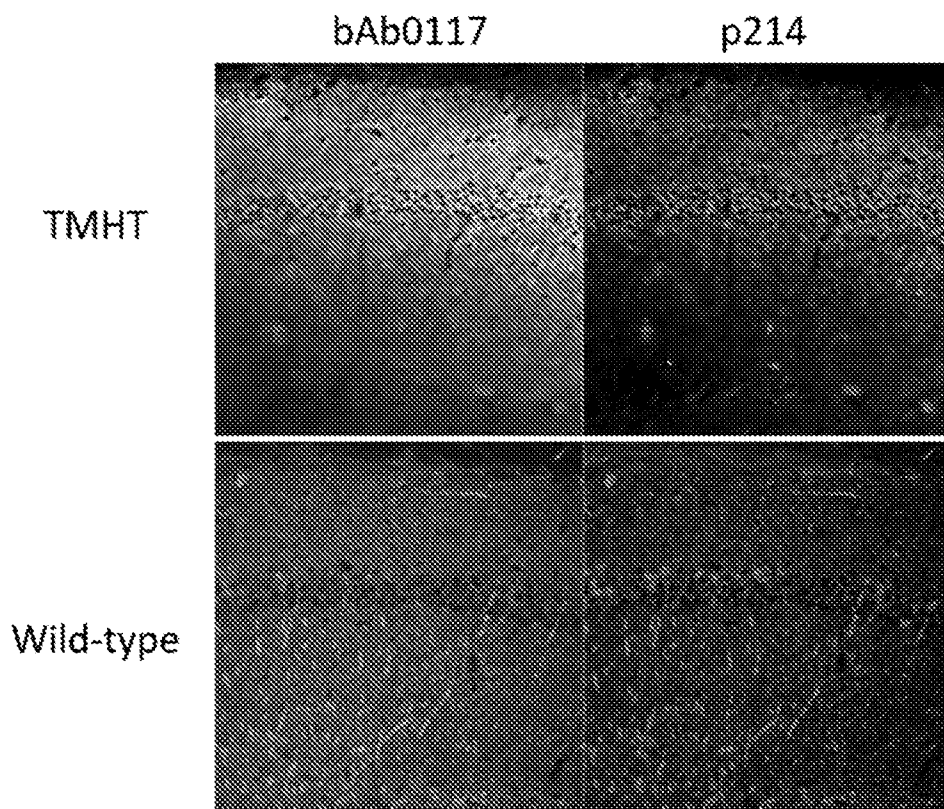


Figure 7

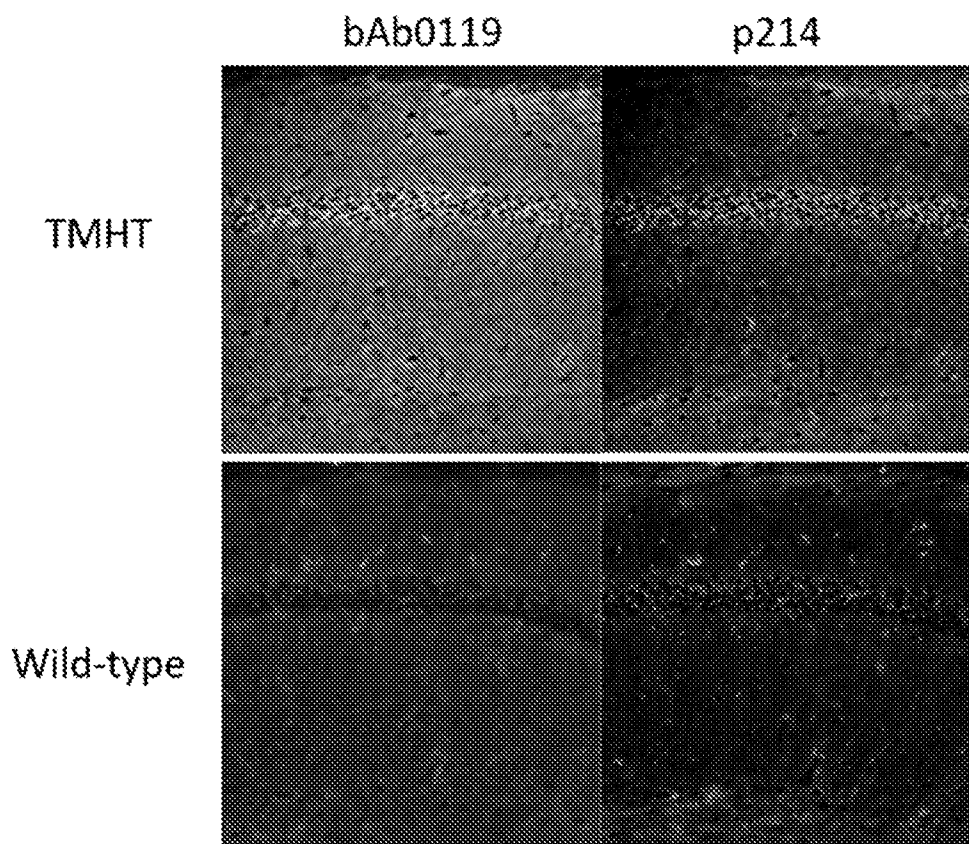


Figure 8

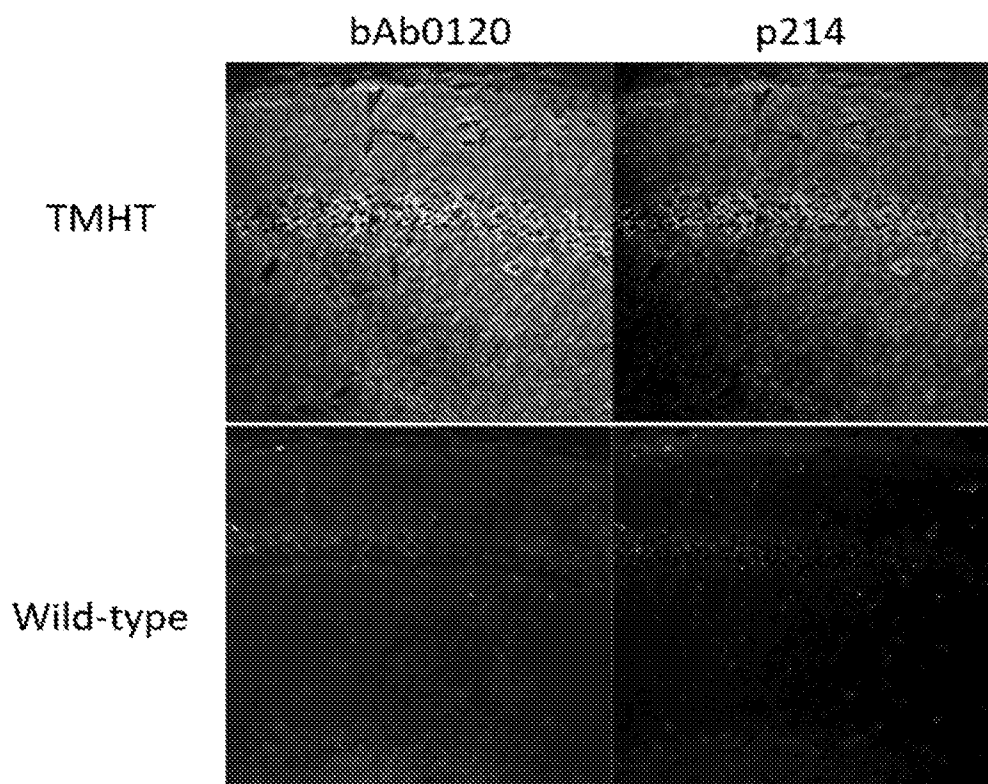


Figure 9.

