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Review Article

Peptide-based brain distribution improvement strategies for nano delivery systems in CNS diseases treatment

Yiran Qian[1,2], Xiaoxi Chang[1], Chenyan Lv[1], Jiachen Zang[1], Guanghua Zhao[1], and Tuo Zhang[1,2] ✉

[1] College of Food Science and Nutritional Engineering, China Agricultural University, Beijing 100083, China
[2] Center of Food Colloids and Delivery for Functionality, College of Food Science and Nutritional Engineering, China Agricultural University, Beijing 100083, China

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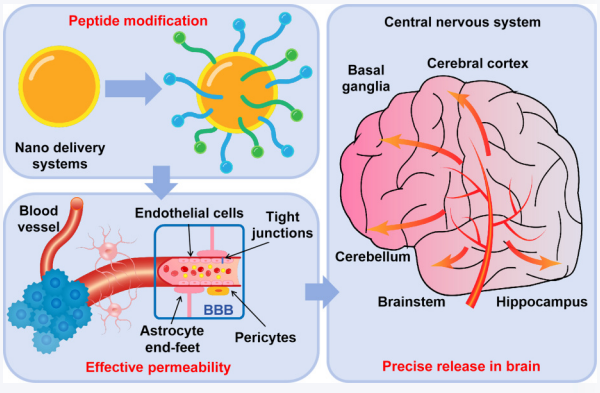
ABSTRACT: Central nervous system disorders constitute a major global public health burden, contributing substantially to morbidity and mortality. Advances in elucidating their underlying pathogenesis have facilitated the approval of an increasing number of proprietary drugs for clinical management of neurological conditions. A critical challenge in drug delivery lies in achieving appropriate tissue distribution, particularly within the brain, where effective therapeutic intervention requires traversing the blood-brain barrier and precisely targeting localized regions. Nanodelivery systems have emerged as a promising approach in biomedicine to address these challenges. Among these, peptides—characterized by their high specificity and relatively small size—are extensively employed to functionalize nanocarriers, thereby enhancing targeted tissue distribution. The conjugation of diverse functional peptides onto heterogeneous nanoscale carriers enables precise, efficient, and multidimensional targeting. This review highlights several representative neurological diseases and systematically discusses strategies for peptide-based functionalization of nanocarriers tailored to these pathological contexts.



KEYWORDS: central nervous system disease, blood brain barrier, cell penetrating peptide, blood-brain barrier (BBB) shuttle peptide, homing peptide

1 Introduction

Diseases of the central nervous system (CNS) affect tens of millions of people and have become one of the most serious medical and public health challenges in modern society. According to estimates by the World Health Organization (WHO), neurodegenerative diseases are projected to surpass cancer as the second leading cause of mortality by 2040 [1]. Most CNS diseases remain incurable, and their pathogenesis has not been fully elucidated. In addition to the lack of effective drugs, the absence of efficient drug delivery methods to the brain represents a significant challenge. The blood-brain barrier (BBB) limits the entry of most drugs, and the complex structure of the brain further complicates the accumulation of drugs through non-invasive administration. To achieve efficient drug distribution in specific brain regions, the development and design of targeted delivery systems are crucial. Nanomaterials are extensively employed in the treatment of CNS diseases to develop universal drug delivery strategies [2]. Their surfaces can present a variety of ligands for precise targeting and controlled release of the drug vector. As research on peptides advances, their role as functional elements has become increasingly prominent. Here, we refer to peptides consisting of 5 to 30 amino acid residues, which may or may not adopt a specific conformation. Several peptides have emerged as promising candidates in the treatment of CNS diseases. Modifications of these peptides have enhanced the blood-brain barrier permeability of nanocarriers and increased their tissue distribution specificity, significantly improving the bioavailability of the encapsulated cargo. This phenomenon is often referred to as the "Trojan Horse" of biology. Nevertheless, CNS diseases are too complex to be addressed by a simple intracerebral delivery vector. Recent trends focus on developing heterogeneous vectors capable of stepwise and multidimensional targeting. This article will therefore introduce the pathological features, effective drugs, and nanomaterial-based treatment strategies for several major CNS

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✉Address correspondence to zhangtuo@cau.edu.cn

diseases, along with a comprehensive summary of peptides used for nanocarrier modification, including cell-penetrating peptides, brain shuttle peptides, and homing peptides (Fig. 1).

2 CNS diseases

The central nervous system comprises the brain, spinal cord, and neural components of the eye. As the principal regulator of vital physiological processes, the CNS is protected by biological barriers and is endowed with a unique immune privilege [3, 4]. Even subtle changes in the microenvironment of the CNS—such as variations in pH, ion strength, or redox status—can have profound impacts on its function [5]. These alterations may result from a variety of factors, including infections (bacterial or viral), metabolic disturbances, ischemia, hemorrhages, inflammation, immune responses, or age-related degeneration, potentially leading to severe neurological disorders. CNS diseases, including stroke, brain cancer, and neurodegenerative diseases, affect billions worldwide, contributing not only to significant patient suffering but also imposing a considerable economic burden on society.

Tissue distribution is a fundamental concept in pharmacology [6], often associated with drug toxicity. The speed and efficiency of drug distribution are critical factors influencing therapeutic efficacy. Specifically, the local concentration of a drug at the target site, along with its duration of action, plays a decisive role in its effectiveness. In clinical practice, intracerebral drug delivery is typically achieved through systemic routes, such as oral administration (non-invasive) and intravenous or intramuscular injection (invasive). Oral administration is commonly preferred due to its convenience and higher patient compliance. However, to achieve therapeutic concentrations of a drug in the bloodstream, higher oral doses are often necessary. Simply increasing the systemic concentration does not always result in higher drug levels in the brain and may even lead to significant adverse effects, such as hepatorenal toxicity, anaphylactic reactions, or worse. These challenges are largely attributed to the brain's protective barriers.

The brain is encased by two distinct barriers: the blood-brain barrier, which separates the brain from the systemic circulation, and the blood-cerebrospinal fluid barrier (BCSFB) [7]. The BCSFB is formed by the choroid and arachnoidal epithelium, providing access to the cerebrospinal fluid (CSF) in the ventricular and subarachnoid spaces, respectively [8]. Like other biological barriers, the BCSFB contains tight junctions, but it has a relatively low electrical resistance (~ 200 Ω·cm^2) and a surface area approximately 1000 times smaller than that of the BBB [9]. Consequently, the BCSFB has received comparatively less attention in the context of CNS drug delivery.

2.1 Blood-brain barrier

The BBB is a physiological structure composed of a layer of tightly packed, metabolically active endothelial cells that regulate the transfer of substances between the blood and neural tissues [10–12]. These endothelial cells, which form the walls of brain capillaries, are the primary constituents of the BBB. They establish tight junctions that restrict paracellular transport. Moreover, these cells contain a variety of cytosolic and extracellular membrane-bound enzymes, which downregulate vesicular transport and efflux pumps. Collectively, these features enable the BBB to play a crucial role in maintaining brain homeostasis.

Although the BBB functions as a formidable barrier, it must allow the passage of essential ions, nutrients, and hormones to support brain function. To achieve this, it employs a series of transport mechanisms. The maximum distance between cells and capillaries is approximately 20 micrometers, a distance that can be crossed by small molecules in less than a second [13]. Certain small hydrophobic compounds (less than 500 Da) can diffuse across the endothelial membrane, while polar molecules such as glucose, amino acids, and some peptides rely on specific transporters that facilitate their entry into the brain's extracellular space. Macromolecules and hormones, on the other hand, are transported via endocytic mechanisms, including receptor-mediated transcytosis (RMT) and adsorptive-mediated transcytosis (AMT) [14]. In the context of CNS diseases, drugs must be delivered directly to the affected areas in sufficient concentrations to provide effective local treatment. However, this delivery process is significantly hindered by the BBB. As a result, the development of

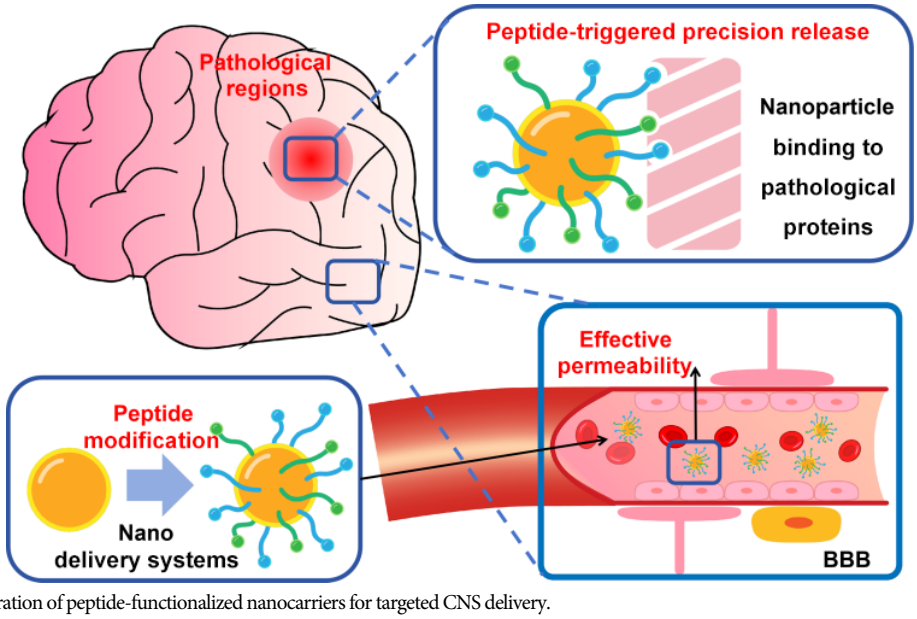

Figure 1 Schematic illustration of peptide-functionalized nanocarriers for targeted CNS delivery.

efficient and non-invasive strategies to enhance drug accumulation in the brain is an urgent need.

2.2 Blood-cerebrospinal fluid barrier

The BCSFB, along with the blood-brain barrier, works to protect the central nervous system. However, the BCSFB is more accessible to potential invaders, resulting in higher permeability of nanomedicines across the BCSFB compared to the BBB. The BCSFB is located in the choroid plexus (CP) of the brain ventricles. The CP is an epithelio-endothelial structure composed of a highly vascularized stroma with connective tissue and epithelial cells [15–17]. The stroma contains fenestrated capillaries with 60 to 80 nm openings, surrounded by connective tissue rich in immune cells. The ventricular side of the stroma is covered by a single layer of cuboidal epithelial cells. These epithelial cells possess the ability to secrete various proteins into the CSF, and could potentially release therapeutic proteins or anti-inflammatory cytokines in response to different neurological conditions. With brisk blood flow (approximately 10 times that of the brain) and highly permeable capillaries, the human CP ensures a high turnover rate of fluid (around 400,000 μL/day), which contains micronutrients, peptides, and hormones necessary for neuronal networks [18]. CSF formation occurs via basolateral carrier-mediated uptake of Na^+, Cl^-, and HCO^{3-}, followed by apical release through ion channel conductance and osmotic water flow through aquaporin 1 (AQP1) channels. Transcellular active transport and secretion in epithelial cells are driven by a dense network of organelles, including mitochondria, endoplasmic reticulum, and Golgi apparatus [19].

However, the ability of the brain to access medication does not correspond to the permeability of the BCSFB. For instance, the movement of proteins and peptides across the BCSFB is highly restricted, primarily occurring through paracellular leaks at incomplete tight junctions and low-capacity transcellular pinocytosis/exocytosis. The pathogenesis and pathophysiology of various diseases are often linked to dysfunctions in the CP and B-CSF barrier. It can be concluded that major alterations involve the production of different proteins by CP epithelial cells, the expression of tight junction (TJ) and transporter proteins, and immune cell trafficking. However, it remains unclear whether the CP responds uniformly to different diseases across all ventricles. A comprehensive understanding of the roles played by the CP in various diseases is crucial, as it may lead to new therapeutic strategies [20].

2.3 Ischemic stroke

Stroke is one of the leading causes of death worldwide and a major contributor to disability due to its high prevalence as a CNS disease. Ischemic stroke (IS) is the most common type of stroke in clinical practice, typically caused by sudden infarction of brain vessels, cardiac arrest, or hypotension [21]. Ischemic stroke generally occurs when blood flow to the brain is obstructed due to thrombosis or ruptured arteries. Disruption of the blood supply results in a lack of energy and metabolic dysfunction, causing tissue damage and widespread neuronal death [22, 23]. Currently, therapeutic approaches for acute IS primarily include thrombolysis, neuroprotection during reperfusion injury, and the restoration of central nervous system cells.

In the event of an IS, the first priority is to clear the blocked vascular area through thrombolysis, which can be achieved via systemic or interventional methods. For patients who are not candidates for endovascular thrombectomy, thrombolysis using drugs remains the only option. Tissue plasminogen activator (tPA) is the only thrombolytic therapy approved by U.S. Food and Drug Administration (FDA), but it has a narrow therapeutic window (3–4.5 h) and is most effective in the early stages of infarction. This limitation is likely due to its short plasma half-life [24]. Additionally, intravenous thrombolysis with Alteplase is currently the only FDA-approved thrombolytic agent [25]. The potential of Tenecteplase to replace Alteplase as a new generation thrombolytic drug has sparked renewed interest in developing new intravenous thrombolytic therapies. In cases of large vessel occlusion, patients may require more potent thrombolytic agents with high efficacy and good safety profiles. Non-immunogenic recombinant staphylokinase and recombinant human prourokinase may fulfill these requirements [26, 27].

To improve and stabilize the effects of thrombolytic therapy, antithrombotic drugs can also be used in combination with intravenous thrombolysis. Short-acting antithrombotic agents such as eptifibatide and argatroban have demonstrated unique potential. Eptifibatide inhibits platelet aggregation via the glycoprotein IIb/IIIa receptor, thereby preventing fibrin crosslinking [28], while argatroban directly binds to thrombin to inhibit fibrin deposition [29].

Although reperfusion of ischemic tissue is crucial for survival, it also triggers oxidative damage, cell death, and aberrant immune responses through the generation of mitochondrial reactive oxygen species (ROS) [30–33]. During this process, neuronal death occurs due to multiple factors, and the resulting damage is largely irreversible. The neuronal death pathways in IS are believed to involve intrinsic and extrinsic apoptosis, ferroptosis, autophagy, necroptosis, parthanatos, pyroptosis, and phagoptosis [22]. Mitochondrial ROS are generally considered a nonspecific response to ischemia, and their clearance represents a critical therapeutic strategy. Antioxidants and anti-inflammatory agents, such as edaravone and fingolimod, have demonstrated effectiveness in stroke models and are currently being evaluated in clinical trials [34]. Additionally, several neuroprotective agents have been developed to mitigate the effects of ischemic tissue reperfusion following thrombolytic therapy. For instance, Otaplimastat, a matrix metalloproteinase (MMP) inhibitor, has been shown to reduce edema and intracerebral hemorrhage in stroke models treated with tPA [35]. Similarly, 3K3A-APC, a PAR1 agonist, reduces neurological injury and promotes vascular integrity [36].

After the initial treatment of IS, the formation of a rich vascular network (angiogenesis) in the damaged brain tissue is crucial for improving patient recovery and facilitating neuronal network regeneration (neurogenesis) of endogenous or transplanted neural stem cells [37–39]. Angiogenesis occurs through three main mechanisms: remodeling of blood vessels to form smaller microvessels, arteriogenesis, and sprouting angiogenesis [40]. Among these, sprouting angiogenesis has been identified as the primary mode of vascular growth following cerebral ischemia [41]. To enhance blood flow in the peri-infarct area, promote blood vessel growth, and restore vascular function, several strategies have been proposed, including the use of endothelial progenitor cells, mesenchymal stem cells, cytokines, growth factors, and non-coding RNAs [42]. Stem cell-based therapies are highly effective in repairing nerve cells, though they typically require invasive drug delivery methods. In contrast, non-invasive delivery of growth factors, cytokines, and angiogenic mediators to the infarcted

peripheral nerve tissue is a more universally applicable direction for current research.

2.4 Alzheimer's disease

As a typical neurodegenerative disease, Alzheimer's disease (AD) has an incidence of approximately 1.5% in individuals aged ≥ 65 years, with the incidence doubling approximately every 5 years of age. In 2019, the global number of people living with dementia was estimated to be 57 million, and this number is projected to reach 153 million by 2050 [43]. The global economic burden of AD and related dementias is also projected to rise significantly, from $4.7 trillion in 2030 to $8.5 trillion in 2040, and up to $16.9 trillion by 2050 [44]. AD is the most common form of dementia, characterized as a progressive, multifactorial neurodegenerative disorder of the central nervous system. Pathologically, it is marked by the presence of senile plaques, neurofibrillary tangles, chronic inflammation, and aberrant neuronal apoptosis [45, 46]. The clinical manifestations primarily include cognitive decline, mental symptoms, behavioral changes, and a gradual loss of daily living abilities.

Despite decades of intensive clinical trials, substantial financial investments, and human capital directed toward AD drug development, many FDA-approved therapeutic drugs remain ineffective in reversing the progression of the disease. These limitations are largely attributed to the complex pathogenesis and intricate brain signaling pathways involved in AD [47]. As a result, AD continues to represent one of the most significant unmet medical needs worldwide. Extensive clinical observations, along with *in vitro* and animal model studies, have revealed numerous molecular alterations and pathological disorders in AD patients and models. These include the well-known overexpression and aggregation of amyloid-beta (Aβ), disturbances in metal ion homeostasis, upregulation of ROS and inflammatory mediators, hyperphosphorylation of the microtubule-associated protein tau, cholinergic dysfunction, and synaptic loss [48–51]. Such complex pathological features have led to the realization that existing single-target drugs are insufficient to treat AD effectively. While the academic community has been exploring multi-target approaches to AD treatment, the unclear etiology of the disease remains a fundamental challenge for researchers. Consequently, personalized precision medicine is now considered essential for the effective treatment of AD [52].

2.5 Parkinson's disease

Parkinson's disease (PD), the second most common neurodegenerative disorder, remains incurable and affects approximately 1% of the population aged 60 and older. In the EUROPARKINSON study, the percentage of undiagnosed screen-detected cases increased from 18% in individuals aged 65–70 years to 36% in those aged 80–85 years [53]. PD is primarily characterized by the degeneration of dopaminergic neurons in the midbrain's substantia nigra (SN) [54], leading to debilitating motor symptoms such as tremors, impaired gait, and speech difficulties, which worsen as the disease progresses [55]. A hallmark pathological feature of PD is the abnormal intra-neuronal deposition of α-synuclein (α-syn), a protein also implicated in other neurodegenerative diseases, including dementia with Lewy bodies (DLB) and multiple system atrophy (MSA) [56].

Among the various fibrillary conformations of α-syn aggregates formed during autocatalytic self-amplification, soluble oligomeric non-fibrillary structures are considered the major toxic species, contributing to neuronal cell death in PD and other synucleinopathies [57, 58]. However, the concentration of α-syn aggregates in the central nervous system is considerably lower compared to other neurodegenerative disease markers such as amyloid-beta (Aβ) or tau aggregates, making it a significant challenge to detect and monitor the onset and progression of PD [59, 60]. Pathological α-syn can propagate to adjacent neurons in anatomically connected brain regions, where it acts as a seed to promote the aggregation and deposition of α-syn monomers [61–63]. This process exacerbates oxidative stress, depolarizes mitochondria, disrupts protein clearance mechanisms, and alters the cytoskeleton. Furthermore, α-syn aggregation in mitochondria generates high concentrations of reactive oxygen species, which further stimulates the expression of inducible nitric oxide synthase (iNOS) and neuroinflammation, leading to neuronal damage and the progression of PD [64].

Various methods have been proposed to slow the progression of PD, primarily focusing on two aspects: targeting specific molecular pathways and specific neuronal rescue pathways [65]. In terms of molecular pathway targeting, α-syn, Leucine-rich repeat kinase 2 (LRRK2), and β-glucocerebrosidase (GBA) are the most commonly implicated pathological features in PD. The aggregation of α-syn has been observed in the majority of PD patients, although it is not the only misfolded protein present in their brains, which is often described as a prion-like spread of Lewy pathology [66, 67]. Therefore, one promising strategy to mitigate cytotoxicity by reducing cellular toxicity and the pathological spread of α-syn is the development of anti-α-syn antibodies or the stimulation of microglia transformation into the M1 phenotype, thereby enhancing immune-mediated clearance of α-syn [68, 69].

The second target, LRRK2, is associated with autosomal-dominant PD, which results in increased LRRK2 kinase activity and subsequent disruption of autophagy [70]. As a result, LRRK2 inhibitors have been developed, and two of these (DNL201 and DNL151) are currently undergoing phase 1B clinical trials. Similarly, targeting GBA is a therapeutic approach based on genetic variations in the GBA gene, which cause mutations in β-glucocerebrosidase. These mutations result in the accumulation of the enzyme in the endoplasmic reticulum, leading to its degradation via the proteasome and reduced lysosomal concentrations. Consequently, lysosomal dysfunction occurs, vesicle transport is impaired, and α-syn aggregation is promoted [71].

On the other hand, neuronal rescue pathway targeting largely focuses on ion metabolism, mitochondrial function, and neuroinflammation. The substantia nigra pars compacta (SNpc) is a typical neuronal population with spontaneous pacemaking properties that regulate calcium ion entry into cells. When calcium absorption is upregulated, oxidative stress, mitochondrial damage, and even cell death may occur [72]. Based on this process, inhibitors of L-type voltage-gated Ca^{2+} channels (Cav1.3) have demonstrated neuroprotective effects in animal models [73]. Additionally, iron overload in the SNpc is a common pathological feature of PD, and research utilizing iron chelation with deferiprone has yielded positive results in PD models [74].

Recent research has focused primarily on mitochondria and neuroinflammation as key therapeutic targets for PD. These two factors are fundamental to the downstream symptoms of PD and are considered the most effective targets for therapeutic intervention. Sargramostim, a recombinant human granulocyte-

macrophage colony-stimulating factor, has shown potential in improving regulatory T-cell function, thereby alleviating PD by reducing inflammation [75]. Similarly, AZD3241, a selective and irreversible myeloperoxidase inhibitor, has been found to suppress microglial activation and protect dopaminergic neurons [76]. The direct involvement of mitochondrial dysfunction in the pathogenesis of PD is well-established [77]. In conclusion, further clinical studies are essential to confirm the therapeutic effects of these interventions on PD. Additionally, the classification and identification of various PD subtypes are critical to advancing the development of targeted treatments for the disease.

2.6 Other CNS diseases

In addition to the previously mentioned diseases, brain cancer, multiple sclerosis (MS), and amyotrophic lateral sclerosis (ALS) represent serious medical challenges, all with extremely low cure rates. In the context of brain cancer, glioblastoma and glioma are the most prevalent and lethal forms. Glioblastoma typically originates in the cerebral hemispheres, with 95% of these tumors arising in the supratentorial region, particularly in the frontal and temporal lobes. These tumors infiltrate the brain parenchyma and generally do not metastasize [78]. The aggressive invasiveness of glioblastoma, coupled with the BBB hindering effective drug delivery, presents a significant therapeutic challenge [79]. Glioma stem cells (GSCs) are a small subpopulation within glioblastomas that exhibit genomic instability, self-renewal, and tumor-initiating properties. These cells are believed to contribute to tumor heterogeneity by differentiating into various glioblastoma subpopulations [80]. Furthermore, GSCs are resistant to apoptosis, regulate the tumor microenvironment, promote angiogenesis, suppress immune responses, and contribute to radiation and chemotherapy resistance [81–84]. The inability of most current treatments to effectively target GSCs is considered a major factor in the recurrence of glioblastoma [84].

MS is a neuroinflammatory disease of the CNS characterized by demyelination and neuronal damage [85]. The annual economic burden of MS in the United States is estimated at approximately US $85.4 billion [86]. While the precise cause of MS remains unknown, the adaptive immune system—particularly T and B lymphocytes—plays a pivotal role in its pathogenesis, driving the development and clinical recurrence of focal lesions [85]. MS lesions, or plaques, result from acute inflammatory damage within the CNS. These lesions typically form around the central vein and are distributed across the CNS, affecting white matter, the cortex, and deep gray matter. During MS relapses, T cells are reactivated by antigen presentation, triggering an inflammatory cascade and recruiting additional immune cells. This response leads to focal edema, myelin destruction, oligodendrocyte damage, and neuronal injury, including axonal damage. However, local inflammation and nerve damage are somewhat sporadic rather than progressive [87]. Disease progression is ultimately driven by the complex interplay between autoimmune responses, neuroinflammation, and neurodegeneration [88].

Amyotrophic lateral sclerosis (ALS) is a devastating neurodegenerative disease characterized by the progressive loss of motor neurons. Although the disease progresses rapidly, it typically spares cognitive function and the sense of self. ALS involves the degeneration of both upper motor neurons (UMNs) in the cerebral cortex and lower motor neurons (LMNs) in the brainstem and spinal cord. This dual degeneration leads to muscle weakness, which gradually evolves into muscle atrophy and paralysis, ultimately resulting in respiratory failure and death [89]. The exact causes of ALS remain unknown. However, several pathogenic mechanisms have been implicated, including disrupted RNA metabolism, glutamate excitotoxicity, protein misfolding and aggregation, endoplasmic reticulum stress, impaired protein and axon transport, oxidative stress, inflammation, and mitochondrial dysfunction [90, 91]. A key feature of ALS is mitochondrial dysfunction triggered by mutations in the SOD1 gene, which exacerbates oxidative stress and contributes to disease progression [92].

3 Peptide-based strategies for CNS disease

Strategies to overcome the BBB can be broadly categorized into those that temporarily disrupt its integrity and those that utilize endogenous transport mechanisms. Among disruptive approaches, transcranial focused ultrasound (FUS) represents one of the most straightforward approaches, which is FDA-approved and clinically implemented [93–95]. FUS-mediated BBB disruption is achieved through the interaction between acoustic waves and intravenously administered cavitation-enhancing agents. Consequently, the implementation of this strategy is demanding in terms of equipment specifications, and the selection of appropriate cavitation-enhancing agents remains a critical consideration [96, 97]. Strictly speaking, FUS constitutes a disruptive strategy for enhancing drug delivery. The potential for FUS to induce structural alterations in the BBB and brain parenchyma necessitates careful consideration. Investigations into FUS-associated biochemical changes have revealed an elevation in a spectrum of pro-inflammatory cytokines, thereby establishing a non-edematous, sterile inflammatory state within the brain [98, 99]. Furthermore, the FUS procedure directly engages neural cells, impacting both the cell membrane and mechanosensitive ion channels, thereby inducing neuronal activity [100, 101]. The potential benefits and adverse effects of this direct neuromodulatory stimulation on neural cells require more comprehensive investigation.

Given the potential risks associated with BBB disruption, significant research efforts are directed towards non-invasive strategies that leverage the BBB's own transport machinery. Within the realm, several delivery vectors possess the capacity to traverse the BBB. Notable examples include therapeutic antibodies, exosomes, and brain-tropic viral vectors, among others [102, 103]. Antibody therapeutics (full-sized immunoglobulins, single-chain variable fragments, antigen-binding fragment, and single-domain antibodies) achieve brain delivery by binding to specific receptors on the BBB, followed by receptor-mediated endocytosis. While this represents a highly precise targeting strategy, the very specificity that enhances BBB permeability concurrently leads to co-localization with lysosomes or receptor-mediated brain clearance. Consequently, excessively high antibody affinity may be counterproductive [104]. Exosomes, particularly those derived from neural or immune cells, possess an inherent brain-targeting capacity, which facilitates their transport across the BBB via receptor-mediated interactions [105]. However, exosomes face substantial challenges in clinical translation. Foremost among these is the difficulty in scaling up production to clinical-grade standards while preserving their purity and functional integrity. Additionally, the lack of a universally accepted medical definition for exosomes hampers the establishment of corresponding regulatory

frameworks. Consequently, significant hurdles must be overcome before exosome-based therapies can be successfully translated to benefit patients [106]. Adeno-associated virus (AAV), an emerging vector for intracranial drug delivery, is characterized by high transduction efficiency, structural stability, low immunogenicity, and minimal toxicity. Nevertheless, engineering modifications are required to confer upon AAVs the capability to traverse the BBB, as discussed in this review.

The complexity of brain diseases makes them resistant to treatment by a single-drug strategy. However, precise and effective drug delivery is essential for alleviating symptoms and reducing patient suffering. Driven by the goal of overcoming these currently incurable diseases, researchers are exploring various strategies focused on stepwise targeting, precision, and responsive release. To achieve these objectives, functional delivery vectors often require the use of nanomaterials with strong modification capabilities. These nanomaterials must feature rigid or semi-rigid structures with multiple surface modification sites or possess inherent receptor-specific binding abilities. Compared to small molecules, the directional modification of these larger "vehicles" to enhance diffusion efficiency presents a greater challenge. Even when nanoparticles exhibit excellent hydrophobicity and optimal shapes, these factors alone do not guarantee effective cellular penetration. To enhance the permeability of bioengineered nanoparticles and enable them to cross biological barriers, peptides are frequently incorporated into delivery systems as efficient ligands.

As research into specific interactions and signal transduction pathways *in vivo* advances, peptides have gained significant attention in the biomedical field. In the body, peptides primarily function as hormones, activating downstream pathways through G protein-coupled receptors to regulate metabolism and various physiological functions [107]. The success of peptides in pharmaceutical applications highlights their potential as specific ligands *in vivo*. Often, peptides need only a specific primary structure to engage and activate their downstream targets. Through interactions with receptors and subsequent induction of endocytosis, peptide-conjugated delivery vectors can be absorbed and transported efficiently. In the treatment of CNS diseases, the coupling of peptides—such as targeting peptides, homing peptides, and shuttle peptides—has emerged as a key strategy. These peptides serve various functions: some facilitate transport, others enhance enrichment, some respond to enzymes or environmental stimuli for controlled release, while others assist with intracellular localization. By modifying or even directly assembling these peptides, complex delivery and release mechanisms can be achieved for designed vectors. This article aims to summarize the various targeting and shuttle peptides applicable to CNS diseases, offering insights for the design of bioengineered nanoparticles for the treatment of these complex conditions.

3.1 Peptide modification

The BBB poses a significant challenge to the entry of most substances with molecular weights greater than 500 Da, particularly in systemic drug delivery [108]. Despite this, the CNS has a continuous and critical need for hormone proteins and other macromolecules, which presents opportunities for targeted drug delivery. For protein and peptide-based drugs, the most efficient strategy for crossing the BBB is to exploit specific receptor interactions through the fusion of peptides. This approach has been successfully applied in various nanoparticulate delivery systems. Ideal carriers for loading and presenting functional peptides include metal-organic frameworks (MOFs), cage-like proteins, exosomes, self-assembling peptides, and polymers, etc. Some peptides are derived from known transport receptors, while others have been identified through phage display technology.

Inspired by chimeric proteins targeting cell receptors, the concept of the BBB shuttle was first proposed by William M. Pardridge in the mid-1980s [109]. Initially, antibodies were explored for receptor-mediated strategies to cross the BBB. IgGs targeting insulin and transferrin receptors showed potential, but their high affinity hindered efficient release into the brain parenchyma [110, 111]. Since then, several receptors located on brain endothelium have been investigated, including apolipoproteins (Apo) A and E [112], receptor-associated protein (RAP) [113], transferrin (Tf) [114], lactotransferrin [115], melanotransferrin (P97) [116], and leptin [117]. One major advantage of using receptors on brain endothelial cells for drug delivery is that endogenous proteins typically exhibit low immunogenicity compared to exogenous analogs or antibodies. Additionally, proteins with quaternary structures offer greater potential for modification and drug loading. However, these advantages are often overshadowed by the growing interest in peptides. Peptides have emerged as a promising alternative due to their near-equivalence to complex proteins in terms of receptor selectivity and binding strength. They combine the low cost of small molecules with the high specificity of biologics, and can be tailored to exhibit either a flexible or rigid structure [111]. Through chemical and genetic engineering, peptides can be conjugated with other proteins or modified to improve blood half-life and optimize binding strength.

In 1999, Stephen Dowdy and his colleagues demonstrated that a fragment of the HIV trans-activator of transcription (TAT) protein could deliver *p*-galactosidase into the brain and other organs [118], marking a pioneering step in the development of BBB shuttle peptides. The TAT family was later recognized as a typical group of cell-penetrating peptides (CPPs), which facilitate the uptake of various molecules, from nanoparticles to small compounds and even large DNA fragments, by cells. However, the delivery of cargoes using CPPs tends to be nonselective. In 2007, the peptide RVG2953 was shown to selectively transport cargo into the brain [119], marking a significant advance in targeted peptide delivery. Additionally, the use of D-enantiopeptides *in vivo* has been demonstrated, offering advantages such as protease stability and reduced immunogenicity compared to their L-peptide counterparts [120]. This milestone marks the beginning of a new era in peptide-based targeting strategies.

Substituting L-amino acids with their D-counterparts in engineered peptide sequences represents one of the most prevalent and effective approaches to enhance *in vivo* stability and prolong circulatory half-life. This strategy capitalizes on the stereoselectivity of endogenous proteolytic enzymes, which primarily recognize and cleave peptide bonds formed between L-configured residues. The distinct stereochemical configuration of D-amino acids renders them resistant to proteolytic degradation by most enzymes. Notably, D-amino acids typically exhibit reduced immunogenicity, consequently mitigating host immune responses upon administration of exogenous therapeutic peptides [121–123]. Peptide chain length constitutes a critical design parameter requiring careful optimization. Excessively short peptides (< 7 AA) often fail to adopt stable amphipathic structures or specific spatial

conformations necessary for productive membrane interactions. Conversely, unduly long peptides (> 30 AA) incur substantially higher synthetic costs while increased steric hindrance may impede effective contact with cellular membranes. Furthermore, extended chains exhibit heightened susceptibility to proteolytic degradation and pose an elevated risk of immunogenicity. Furthermore, peptide cyclization represents a crucial strategy for enhancing bioactivity, typically achieved through the introduction of disulfide bonds. Cyclic peptides exhibit significant improvements in stability, half-life, and membrane permeability. Moreover, in certain cases, cyclization can further augment affinity towards their recognition sites [124–127]. Numerous similar structure-activity relationship (SAR) optimization strategies exist, while our focus centers on the primary structure of the peptides themselves, which we will systematically categorize and summarize (Fig. 2).

3.2 Cell penetrating peptides for BBB crossing

Cell-penetrating peptides are short peptides with amphiphilic and polycationic properties, widely used to enhance cell absorption and transport efficiency [128–130]. They are particularly useful for intracellular delivery, where CPPs facilitate lysosomal escape to enable the release of cargo inside the cell. Some CPPs, like TAT, are also employed to penetrate the BBB. However, a single CPP modification often does not suffice for effective BBB penetration. Therefore, CPPs are typically used in conjunction with other modifications to improve the bioavailability of drug delivery vectors. CPPs were initially inspired by viral proteins from influenza, HIV, and other specific viruses. In fact, some viral carriers were originally utilized for drug delivery, demonstrating unexpected efficacy in crossing both extracellular and intracellular barriers. However, the immunogenicity associated with viral vectors remains a significant challenge. As a result, researchers have isolated tissue-penetrating motifs to design and modify non-viral carriers [131].

The exact mechanism by which CPPs facilitate the penetration of biological barriers, such as the BBB, remains unclear. It is believed that the amphiphilic structure of CPPs allows them to insert into lipid bilayers, such as cell membranes, triggering endocytosis of the entire nanoparticle-based delivery system. Some hypotheses suggest that these motifs induce membrane invagination, leading to endocytosis [132]. Several viral-derived motifs have been studied for their ability to penetrate endosomal membranes, including those from the influenza virus (e.g., GALA, KALA, RALA, E5, K5, H5WYG, JST1, influenza virus hemagglutinin-2 analogue (INF7), and CADY) and HIV (e.g., glycoprotein 41 (Gp41), TAT, fusogenic peptide 23 (FP23), HIV gag peptide (HGP), and PEP1) [133–138]. These motifs show promise for use in intracerebral drug delivery.

Many CPPs have been shown to successfully deliver drugs to the CNS by crossing the BBB. The transmembrane transport facilitated by CPPs typically involves non-specific interactions. These peptides are largely derived from phage library screenings, virus-specific motifs, and naturally occurring peptides, such as antimicrobial peptides and neurotoxins. CPPs can effectively mediate the internalization of a wide range of substances, including small molecules, exogenous proteins, plasmid DNA, antigens, fluorophores, and peptide nucleic acids into the cytoplasm and nucleus of cells [139–144]. In addition to their diverse cargo capacity, CPPs share several common characteristics: rapid cell membrane penetration, high levels of nuclear accumulation, consistent uptake efficiency at both 4 and 37 °C, and low sensitivity to inhibitors of clathrin-mediated endocytosis [145]. Table 1 presents a summary of representative CPPs, providing insights for the design and modification of nanocarriers for intracerebral drug delivery.

To date, the TAT family and poly-Arg peptides remain among the most commonly used CPPs. However, these peptide motifs lack sufficient selectivity for the brain. While they can enhance the permeability of carrier molecules in tissues and cells, this improvement in permeability does not always translate into increased accumulation of cargo in the brain. In fact, it is insufficient for a small number of drugs to diffuse into the cerebrospinal fluid. Qualitative and quantitative evidence supporting CPP-induced BBB penetration is limited. Most studies rely on fluorescence-labeled visualization techniques to track and semi-quantify the distribution of delivery vectors and their cargo within brain tissue. While some studies have demonstrated rapid accumulation of vectors in the brain and improved therapeutic effectiveness, other research suggests that these CPPs and their

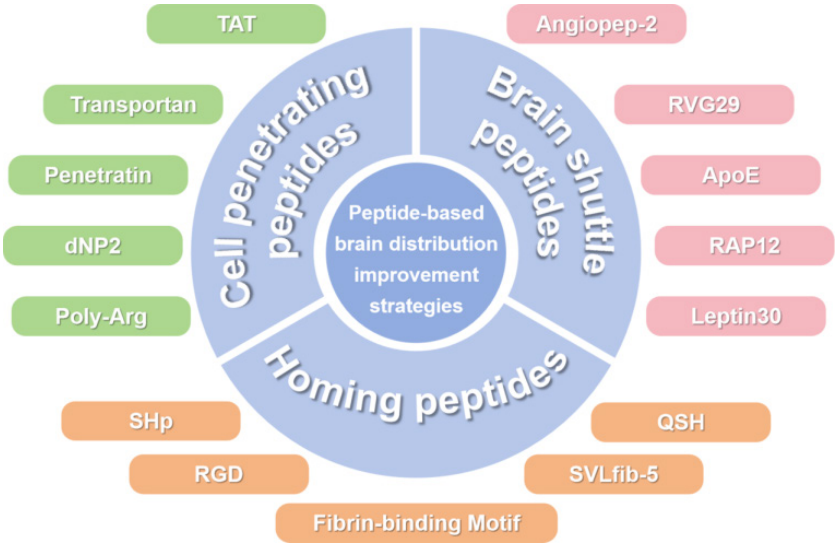

Figure 2 Schematic overview of peptide modification strategies for nanocarriers.

Table 1 Typical CPPs that applied for crossing the blood-brain barrier

Peptide	Sequence	Cargoes	Diseases	Ref.
Poly-Arg	RRRRRRRR	Docetaxel	Glioblastoma/Glioma	[146]
TAT	CAYGRKKRRQRRR	Adriamycin (DOX)	Glioblastoma/Glioma	[147]
Transportan	GWTLNSAGYLLGKINLKALAALA KKIL	siRNA	Glioblastoma/Glioma	[148]
Penetratin	RQIKIWFQNRRMKWKK	Leptin	Obesity	[149]
dNP2	KIKKVKKKGRKKIKKVKKKGRK	Cytotoxic T-lymphocyte antigen 4	Multiple sclerosis	[150]
Cell-penetrating artificial mitochondria-targeting peptide (CAMP)	YGRKKRRQRRRLLRAALRK	Human metallothionein 1A	Parkinson's disease	[151]
Astrotactin 1-derived peptide (AP)	RRRWCKRRR	Cytoplasmic tail of Cytotoxic T-Lymphocyte Antigen-4 (ctCTLA)	Autoimmune encephalomyelitis	[152]
Tachyplesin	RFCWKVCYKGICFKKCK	Anti-miR-210	Glioblastoma/Glioma	[153]
Model amphipathic peptide (MAP)	KLALKLALKALKAALKLA	Tacrine	Alzheimer's disease	[154]
B6	CGHKAKGPRK	Curcumin	Alzheimer's disease	[155]
Bax-binding domain of Ku70 protein	QLPVM	DOX, Erlo	Glioblastoma/Glioma	[156]
Transportan 10 (TP10)	AGYLLGKINLKALAALAKKIL	Dopamine	Parkinson's disease	[157]
Pep-1	KETWWETWWTEWSQPKKKRKV	Phosphatidylethanolamine-binding protein 1	Ischemia-induced inflammation and neuronal damage	[158]
Histidine-rich amphipathic peptide (LAH4)	KKALLALALHHLAHLALHLALAL KKAC	Adeno-associated virus (AAV)	/	[159]
SynB3	RRLSYSRRRF	Endomorphin	/	[160]
Mel	GIGAVLKVLTTGLPALISWIKRKR QQ	pDNA	/	[161]
Kaposi fibroblast growth factor (kFGF)	AAVALLPAVLLALLAP	pDNA	/	[161]
PasR8	FFLIPKGRRRRRRRRGC	pDNA	/	[161]
Antennapedia	RQIKIWFQNRRMKWKK	DNA	/	[162]
AAV.CPP.16	TVSALK	AAV	/	[163]
AAV.CPP.21	TVSALFK	AAV	/	[163]
PepH3	AGILKRW	/	/	[164]
PepNeg	SGTQEEY	/	/	[165]
apidaecin	ONNRPVYIPRPRPPHPRL	/	/	[166]
oncocin	VDKPPYLPRPRPPRRIYNR	/	/	[166]
Apanin	CNCKAPETALCARRCQQH	/	/	[167]
HAI peptide	HAIYPRH	/	/	[168]
Penetratin derivative (DK17)	DRQIKIWFQNRRMKWKK	/	/	[169]

conjugated structures may become trapped within the brain endothelium [14]. In recent years, despite the promise of novel delivery strategies, the contribution of CPP conjugation to improving the distribution of delivery vectors in the brain appears to be minimal.

3.3 Peptides for receptor-mediated BBB transport

Most BBB shuttle peptides that interact with transporters are derived from either neurotropic biomolecules or phage display biopanning. These peptides can be classified into endogenous and exogenous categories [170]. Endogenous motifs typically include hormones, specific protein domains, and lipoproteins, while exogenous motifs may include viruses and neurotoxins. Receptor-mediated BBB shuttle peptides serve as key modifications to drug delivery systems, conferring selective absorption properties to nanoparticles that would otherwise be blocked by the BBB [171]. When screening for receptor-mediated BBB shuttle peptides, several principles must be carefully considered: (1) The target receptor must be expressed at higher levels in cerebral vascular endothelial cells compared to most other cells; (2) receptor-mediated endocytosis must lead to the eventual exocytosis or release of the vesicle on the opposite side of the endothelial barrier; (3) the shuttle peptide should be recognized by the target receptor based on its primary structure, rather than its higher-order structure; (4) the transporters should exhibit high turnover efficiency, with stable function and localization in epithelial cells, regardless of the cell's state or microenvironment.

When discussing BBB transport receptors, transferrin receptor 1 (TfR1) and low-density lipoprotein receptors (LDLRs) are of primary importance. TfR1 is a key component of the pathway responsible for mediating iron ion uptake into cells and plays a crucial role in regulating cellular iron metabolism and maintaining iron balance [172]. It has also gained attention in cancer treatment, as cancer cells require large amounts of iron to fuel rapid

proliferation, leading to significant upregulation of TfR1 on the cell surface [173]. TfR1 facilitates iron uptake by binding to the iron-carrying protein transferrin. Structurally, TfR1 is formed by the cross-linking of two homologous dimers via disulfide bonds. Each monomer contains a large extracellular C-terminal region, a single transmembrane region, and a short N-terminal region. The C-terminal region, which is the outer functional domain, includes binding sites for transferrin. In addition to transferrin, ferritin has also been shown to cross the BBB via TfR1 [174]. Several peptide sequences, such as T7, have been demonstrated to interact with TfR1, facilitating specific absorption and transport across the BBB [175].

In contrast, the LDLR family is more diverse than TfR1. Members of the LDLR family share common structural motifs, including LDLR type A repeats (which mediate ligand binding), epidermal growth factor (EGF)-like domains (which are involved in pH-dependent ligand release in endosomes), transmembrane anchors, and cytoplasmic domains [176]. Additionally, LDLR, VLDLR, and LRP8 (ApoER2) possess O-linked sugar domains outside the plasma membrane and NPxY motifs in the cytoplasmic domain. LRP1 and LRP2 have large extracellular domains, while LRP5/6 contains a PPPSP motif in the cytoplasmic domain [177]. LDLR, in particular, is commonly used in peptide recognition applications. Its natural ligands, such as ApoB, ApoE fragments, and angiopep-2, are widely exploited in modifying intracerebral delivery vectors, along with peptides like Pep-22 derived from phage libraries. A summary of typical transport-binding peptides, along with some peptides that cross the BBB through unknown receptors, is provided in Table 2.

3.4 Homing peptide for precision release in brain

The brain is an incredibly complex organ, with many functional areas and compartments. Even if a drug successfully crosses the blood-brain barrier and enters the brain, additional design considerations are required to ensure it interacts effectively with the targeted diseased areas or receptors. In peptide-based drug delivery strategies, homing peptides — artificially selected peptide motifs — can be used to specifically interact with pathological regions in CNS diseases. By targeting these regions, nano delivery vectors can be directed to the disease site in the brain with enhanced efficiency. Since the development of phage library screening techniques, a large number of homing peptides have been identified. There are several classic examples of homing peptides being applied in the treatment of CNS diseases.

For example, in Wang's study, the fibrin-binding motif Cys–Arg–Glu–Lys–Ala (CREKA) was fused with a self-assembling nanoparticle, which presented the peptide as an antigen on its surface [196]. The fibrin-binding peptide specifically binds to thrombus regions in the brain, allowing the attached thrombolytic peptide to be delivered directly to the lesion site, facilitating thrombus degradation. This homing peptide has been widely used in the design of delivery vectors for thrombolytic therapies *in vivo*. In a separate study by Hong, the peptide "CLEVSRKNC" derived from phage display was shown to selectively home to ischemic stroke tissue, without binding to non-ischemic brain areas [197]. Unlike the fibrin-binding peptide, this homing peptide co-localized

Table 2 Typical peptides that applied for crossing the blood-brain barrier by transporters[a]

Peptide	Sequence	Transporter	Diseases	Ref.
Angiopep-2	TFFYGGSRGKRNNFKTEEY	Low density lipoprotein receptor related protein 1	Glioblastoma/Glioma	[178]
RAP12	EAKIEKHNHYQK	Low density lipoprotein receptor related protein 1	Glioblastoma/Glioma	[179]
RVG29	YTIWMPENPRPGTPCDIFTNSRGKRASNGGGG	Nicotinic acetylcholine receptor	Glioblastoma/Glioma	[180]
RVG15	YTIWCDIFTDSRQKR	Nicotinic acetylcholine receptor	Glioblastoma/Glioma	[181]
ApoE	LRKLRKRLLR	Low density lipoprotein receptor	Glioblastoma/Glioma	[182]
ApoB	QSDIVAHLL	Low density lipoprotein receptor	Alzheimer's disease	[183]
Aβ peptide	KLVFFAED	Receptor for advanced glycation end-products	Alzheimer's disease	[184]
DBP (VP35)	PKTRNSQTQTD	Light chain DYNLL1 (LC8) of dynein	/	[185]
Leptin30	YQQILTSMPSRNVIQISNDLENLRDLLHVL	Leptin receptor	/	[186]
T7	HAIYPRH	Tranferrin receptor-1	Glioblastoma/Glioma	[175]
B6	GHKAKGPRK	Tranferrin receptor-1	Glioblastoma/Glioma	[175]
T12	THRPPMWSPVWP	Tranferrin receptor-1	Glioblastoma/Glioma	[175]
G23	HLNILSTLWKYR	Ganglioside GM1	Glioblastoma/Glioma	[187]
Pep-22	NH2-C6-[cMPRLRGC]c-NH2	Low density lipoprotein receptor	Glioblastoma/Glioma	[188]
Transferrin receptor-binding peptide (THR)	RMAPEEIIMDRPFLYVVR	Tranferrin Receptor-1	Spinal muscular atrophy	[189]
CRT peptide	CRTIGPSVC	Tranferrin Receptor-1	Alzheimer's disease	[190]
TGN peptide	TGNYKALHPHNG	Unknown	Alzheimer's disease	[191]
g7	GFtGFLS(O-beta-D-glucose)-NH2	Unknown	Huntington's disease	[192]
9D-RDP215	H-fWrIrIrrPrrIrIrWf-NH2	Unknown	Glioblastoma/Glioma	[193]
Pep-3	CNSHTQGKC	Unknown	/	[194]
Ppe-9	CLWRPAADC	Unknown	/	[194]
/	GLHTSATNLYLH	Unknown	/	[195]
/	VAARTGEIYVPW	unknown	/	[195]

[a]The lower case letters represent the right-handed conformation of the amino acid.

with neuronal cells undergoing apoptosis in the penumbra region of the stroke lesion. As a result, this homing peptide has been widely applied in the treatment of ischemic stroke [198–200].

In the treatment of Alzheimer's disease, the accumulation and deposition of amyloid-beta (Aβ) protein, along with the aggregation and hyperphosphorylation of tau, are key pathological features [201]. Therefore, targeting Aβ or tau is critical for improving the therapeutic efficacy in Alzheimer's disease. In fact, many of the peptides designed for binding to pathological proteins in Alzheimer's disease not only target these proteins but also have the ability to affect, inhibit, or even reverse their aggregation and pathological conformation [202, 203]. For example, in Tim's study, the mirror image of tau (271–316) was used as a target for phage display selection and next-generation sequencing (NGS) to analyze the selection results [204]. The study showed that the enriched peptides could effectively stop tau aggregation *in vitro* and reduce tau fibril formation in HEK293T cells. A similar strategy was employed for the treatment of Parkinson's disease. In Gyeongji's study, a small 30-amino-acid peptide derived from the C-terminal domain of α-synuclein was used to specifically bind to synaptobrevin-2 (Syb2) [205]. This α-synuclein-derived peptide motif competes with α-synuclein oligomers for binding to Syb2 and significantly restores soluble N-ethylmaleimide-sensitive fusion protein attachment protein receptor (SNARE)-mediated vesicle fusion in the presence of the α-synuclein oligomers. Table 3 summarizes several homing or targeting peptide sequences that have been used in the treatment of typical CNS diseases.

3.5 Clinical translation gaps of peptide-modified nanocarriers

Peptide-based modification of nanocarriers has emerged as a predominant research strategy for direct and efficient enhancement of brain drug distribution. Nevertheless, the influence of peptide functionalization on critical system attributes—including versatility, immunogenicity, biodegradability, toxicity, and long-term safety—remains inadequately characterized. Moreover, quantitative assessment of the precise contribution of peptides to enhanced

Table 3 Typical peptides for precision release in brain[a]

Peptide	Sequence	Target or receptor	Diseases	Ref.
Fibrin-binding motif	CREKA	Fibrin	Ischemic stroke	[206]
SHp	CLEVSRKNC	Neuronal cells undergoing apoptosis at the penumbra region of stroke lesions	Ischemic stroke	[200]
CooP	CGLSGLGVA	Fatty acid binding protein 3 (FABP3)	Glioblastoma/Glioma	[207]
/	NIDPNAV	Lysophosphatidylcholine (LPC) induced demyelination setting	Glioblastoma/Glioma	[208]
Harakiri-19 (HRK-19)	HAVRNGRRGDGGAVPIAQK	Tumor cells	Glioblastoma/Glioma	[209]
Pep1-L	CGEMGWVRC	IL13Rα2	Glioblastoma/Glioma	[210]
AP-1	CRKRLDRNC	IL-4R	Glioblastoma/Glioma	[211]
RGD	CRGDC	Integrin $\alpha v\beta 3$, $\alpha v\beta 5$ and $\alpha v\beta 1$	Glioblastoma/Glioma	[212]
SVLfib-5	DWGVGGMWPDADAWPQ	Aggregated α-syn	Parkinson's disease	[213]
C-terminal fragment of α-amyloid protein 1 (CFα1)	TVEGAGSIAAATGFVKKDQLGKNEEGAPQE	Synaptobrevin-2	Parkinson's disease	[205]
CFα2	KKDQLGKNEEGAPQEGILEDMPVDPDNEAY	Synaptobrevin-2	Parkinson's disease	[205]
αsyn-pep1	NH_2-KVWDLKPFWEVG-COOH	α-synuclein	Parkinson's disease	[214]
αsyn-pep2	NH_2-FDFSSIIQHKTR-COOH	α-synuclein	Parkinson's disease	[214]
αsyn-pep4	NH_2-DDLHSRRIGVTT-COOH	α-synuclein	Parkinson's disease	[214]
αsyn-pep5	NH_2-NLDSTLTLRLFV-COOH	α-synuclein	Parkinson's disease	[214]
/	LIAIMA	Aβ1_42	Alzheimer's disease	[215]
QSH peptide	QSHYRHISPAQV	Aβ plaques	Alzheimer's disease	[216]
αRAP peptide	FAEKFKEAVKDYFAKFWDGSGELKVLMEKEL	The receptor for advanced glycation endproducts (RAGE)	Alzheimer's disease	[217]
/	KLVFF	Full-length Aβ	Alzheimer's disease	[218]
/	PYRWQLWWHNWS	$A\beta_{1-10}$	Alzheimer's disease	[219]
D3	[D]DRPRTRLHTHRNR	Amyloid plaques	Alzheimer's disease	[220]
Tet-1	NH2-HLNILSTLWKYR-COOH	Trisialoganglioside clostridial toxin receptors (GT1B)	Alzheimer's disease	[221]
/	LPFFN	$A\beta_{40}$	Alzheimer's disease	[222]
Aβ oligomer epitope peptide 2 (AOEP2)	FDYKAEFMPWDT	Aβ oligomers	Alzheimer's disease	[223]
/	[D]KLVFW-(Aib)	$A\beta_{42}$	Alzheimer's disease	[224]
p-NH	[D]NITMNSRRRRNH	PHF6 fibrils	Alzheimer's disease	[225]
TD28	[D]TTSLQMRLYYPP	Tau3RD, full lengths tau fibrils	Alzheimer's disease	[226]

[a] [D] represents the right-handed conformation of the amino acid.

brain delivery efficiency is frequently lacking. The pharmacological complexity of nanodelivery systems is further amplified by intricate peptide modifications, presenting several key challenges:

(1) Species-dependent receptor heterogeneity: Significant interspecies variations exist in receptor abundance (e.g., TfR, InsR, insulin-like growth factor 1 receptor (IGF1R), low-density lipoprotein receptor-related protein 1 (LRP1)) between human and murine brain capillaries, limiting the translational relevance of preclinical data [227].

(2) Limitations of healthy models: Biodistribution and pharmacokinetic (PK) studies conducted in healthy animal models inherently fail to recapitulate pathological conditions; PK properties may undergo significant alterations across different disease stages. Consequently, disease-specific models are essential for accurate evaluation of delivery efficiency.

(3) Off-target binding: High expression of target receptors in peripheral tissues can lead to unintended accumulation and reduced brain specificity.

(4) Protein corona interference: Interaction with complex blood components forms a protein corona on the nanocarrier surface, potentially obscuring the functional activity of conjugated peptides.

In summary, the nanomedicine platforms discussed in this review currently demonstrate suboptimal clinical translation outcomes. Substantial refinement and comprehensive characterization through further fundamental research remain imperative to realize their therapeutic potential.

4 Peptide modification for nano delivery systems

For most CNS diseases, achieving a complete cure or disease reversal remains largely elusive, with clinical trials still lacking conclusive evidence of success. In this context, the field of nanobiotechnology — particularly the design of nanocarriers — has gained significant attention. As discussed earlier, the modification of peptides plays a pivotal role in the development of nanodelivery systems, especially in enhancing the efficient penetration of the BBB, a critical hurdle in treating CNS disorders. Whether utilizing exosomes, liposomes, cage proteins, peptide self-assembled nanoparticles, polymers, or micelles, the effective delivery of therapeutic agents to the brain often hinges on incorporating peptides that facilitate BBB penetration.

In practical applications, transport efficiency across the BBB is typically enhanced by peptides that mediate receptor-mediated transcytosis, often supplemented by CPPs. To further enhance targeting to specific pathological areas, additional peptides are incorporated to improve the nanocarrier's accumulation at the disease site. This strategic modification not only improves the distribution of the delivery vector within the tissue but also optimizes brain-specific delivery, ultimately boosting the bioavailability of therapeutic agents.

In delivery studies targeting several representative CNS disorders, the selection of peptide modifications often lacks strong disease-specificity. In reality, however, peptide selection can be more strategically tailored based on disease progression, severity, and patient physiological status [228]. For instance, different CNS diseases inevitably exert distinct influences on the protein expression profile of the BBB. Leveraging proteomics research, the expression patterns of endothelial cell receptors can be analyzed to inform the tailored design of delivery vectors. Furthermore, these disorders are frequently characterized by significant inflammatory responses, which can impair the functionality of specific pathways. Consequently, robust experimental validation of the *in vivo* delivery efficiency becomes essential.

Current research indicates that the onset of several representative CNS disorders is associated with alterations in the expression levels of specific receptors. To summarize concisely: In Alzheimer's disease patients, the BBB exhibits downregulated expression of GLUT1 [229], LRP-1 [230], and P-glycoprotein (P-gp) [231], alongside upregulated expression of RAGE [230]. Furthermore, significant deficiencies are observed in insulin receptor (InsR) and insulin-like growth factor-1 receptor (IGF-1R) expression [232]. Conversely, Parkinson's disease patients exhibit downregulated expression of GLUT1, L-type amino acid transporter 1 (LAT-1) [233], nicotinic acetylcholine receptors (nAChR) [234], and P-gp (late stages). In contrast, P-gp expression is upregulated during early stages of the disease [235], alongside increased expression of intercellular adhesion molecule 1 (ICAM-1) [236]. In brain tumor patients, the BBB exhibits upregulated expression of ATP-binding cassette (ABC) efflux transporters [237], TfR [238], LRP-1 [239], and ICAM-1 [236], alongside downregulated expression of major facilitator superfamily domain-containing protein 2a (Mfsd2a) [240]. Likewise, stroke patients exhibit upregulated expression of TfR and ICAM-1 [236, 241].

Therefore, the rational selection and application of peptide-based targeting strategies must be fundamentally informed by the distinct molecular signature of the BBB in each disease context. It is against this backdrop of understanding disease-specific BBB biology that we examine the following classic design paradigms for peptide-modified nanocarriers. In this section, we will explore several classic strategies for designing peptide-based nanocarriers, with a particular focus on their application in treating typical CNS diseases.

4.1 Ischemic stroke

Based on the therapeutic methods mentioned above, a number of nanodelivery systems have been proposed for the treatment of IS. The most commonly used peptides include two types, one targeting fibrin, and one homing peptide targeting necrotic nerve tissue after stroke.

In the research of Lu et al., Rapamycin is encapsulated in self-assembled micelles consisted of ROS-responsive and fibrin-binding polymers [21]. A special fibrin-binding peptide (CREKA) was combined together to facilitate micelle retention in the ischemic brain areas. While improving drug tissue distribution, antioxidation, autophagic neuron survival and microglia modulation were realized in a combined treatment. Dong et al. designed a bionic drug delivery system with multi-step targeting capability, namely SHp-NM@Edv/RCD (SNM-NPs) [200]. Modified by stroke homing peptide (SHp), SNM-NPs can target damaged neurons more quickly, 5.68 times more efficiently than β-cyclodextrins (RCD). The RCD wrapped in SNM-NPs then responds to reactive oxygen species, leading to the release of Edaravone (Edv), clearing ROS, inhibiting neuroinflammation, and reducing neuronal apoptosis by 90%. Similar strategy appears in Yang et al.'s design [242]. ROS response nanocarriers constructed with cyclodextrin-derived materials modified by SHp showed more efficient distribution in the damaged brain of ischemic stroke mice, and showed very significant localization in endothelial cells and neurons. Zhang et al. constructed a shear and ROS dual response system (UK@Fuc/CDPC-PTPCS) for the sequential targeted delivery of the thrombolytic urokinase (UK) and the

neuroprotective drug cytoplasmic choline (CDPC) [243]. Similarly, ischemic neurons were actively identified by modification of SHp.

A good example of a stepwise target belongs to Wang's work (Fig. 3) [196]. Thrombus targeting peptide, stroke targeting peptide and BBB penetrating peptide were fused together, constructing a nanoparticle based on self-assembling peptides. By crossing the blood-brain barrier and targeting focal areas in the brain, thrombolytic peptides and antioxidant MnO_2 nanozyme are delivered to key sites in a synergistic way.

A recent article mentioning the concept of stepp-wised targeting in the treatment of IS comes from Li et al. [244]. The main focus of this work is the excess ROS produced by ischemia-reperfusion injury and the neuroinflammatory response. BBB penetrating peptide (ANG) and SHp were fused on the DSPE-PEG, respectively, and simultaneously presented on the nanoparticle through the self-assembly of the polymer. When the nanocarriers reach the lesion, the mercaptosterone structures are exposed to high levels of ROS, causing the breakdown of NPs, clearance of ROS and release of Danshensu (salvianic acid A, DSS) via glial cells.

4.2 Alzheimer's disease

Up to now, the application of antibodies to achieve fundamental treatment is considered to be the hope of the next generation of AD therapies, because antibodies-based drugs can directly target its pathology and may block the aggregation process of Aβ monomers to toxic oligomers [245]. Monoclonal antibody targeting aggregated Aβ and plaques unexpectedly received approval in 2021 for AD treatment, namely Aducanumab. This was the first approval of AD drug since 2003 [246]. Lecanemab is also an Aβ targeting antibody approved by the FDA in 2023. Actually, these candidates have shown remarkable results in animal experiments, but have always failed in large-scale human trials. On the whole, the treatment of AD requires a typical multi-target therapy mode, but small molecular or antibodies drugs usually follows the "one molecule one target" pattern. Besides, the access of antibody to the CNS has not been properly resolved.

In order to realize multi-target comprehensive treatment of AD, it is necessary to introduce nano system composed of varied

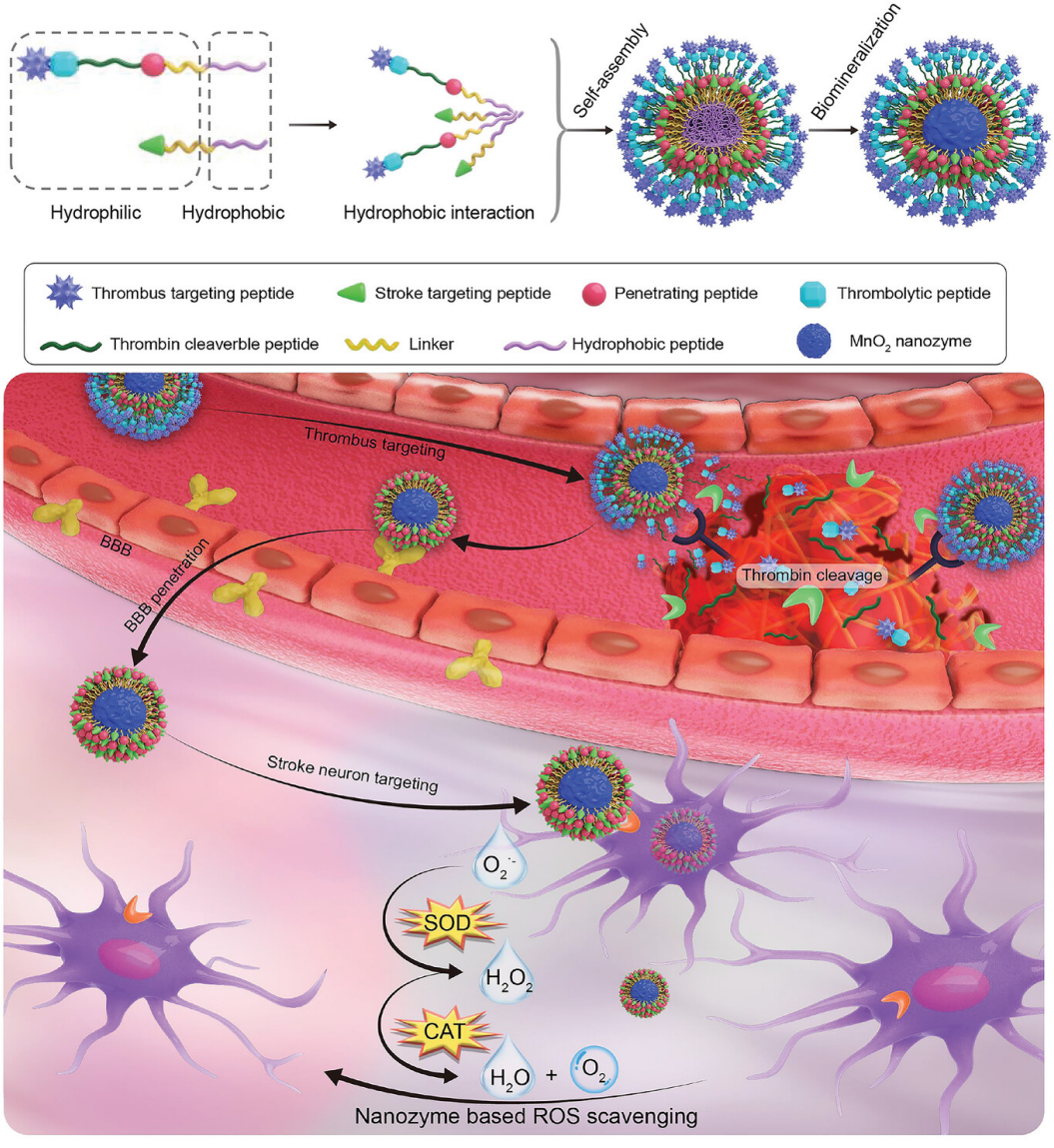

Figure 3 Schematic illustration of the biomineralization synthesis of PNzyme/MnO_2 and its application as a smart multifunctional therapeutic for the treatment of IS. Reproduced with permission from Ref. [196], © Wiley-VCH GmbH 2023.

functional components. Although this approach has not been significantly advanced in the biomedical community, numerous novel design strategies and delivery systems have been elucidated and have achieved remarkable results in animal studies. In addition to the design of multi-functional delivery vectors, the permeability and absorption efficiency of brain are also aspects that must be considered, especially the penetration efficiency of the BBB. In the treatment of AD, nanoparticles cross the BBB in a way that varies little from other CNS disorders. One of the most widely used and effective approaches is the delivery of nanoparticles through receptor-mediated endocytosis. Wang and coworkers modified angiopep-2 to the surface of liposome nano drug delivery system, achieving an effective BBB penetration effect [247]. Icariin (ICA) and tanshinone IIA (TSIIA) loaded in the liposome inhibited neuroinflammation and oxidative stress, reduced apoptosis, protected neurons and improved cognitive function of AD mice. Liron et al. constructed a β-Amyloid targeting nanodrug for the delivery of nucleic acids [248]. D peptides (D1 or D3) were conjugated to the poly(β-L-malic acid-trileucine)-copolymer. The results demonstrated that D3-conjugated nanopolymer drug is able to achieve neuron-selective delivery of miRNA. In Yang's strategies, a multifunctional peptide was used as structural basis for targeting [249]. The peptide VLC was generated by the fusion of the vascular cell adhesion molecule-1 (VCAM-1) high-affinity peptide VHS25 and the neuroprotective ApoE-mimetic peptide COG1410 (COG). The VLC-peptide based NPs effectively homed to pericyte lesions and released curcumin upon ROS stimulation, and the neurovascular function of AD mice was improved by forming a positive feedback loop.

Interestingly, in the research of Liu et al., a desighed peptide (KLVFFAED, Aβ peptide) was fused on a peptide delivery system, inspired by the transportation of Aβ in brain (Fig. 4) [250]. Based on the past research, Aβ could be easily internalized into the brain through RAGE, which is highly expressed and closely related to the progression of AD. The loading of Aβ peptide not only makes an excellent contribution to the BBB penetration of the delivery system, but also can specifically target AD lesion areas in the brain. Through the modification of a peptide, multi-step targeting can be completed, and the delivery efficiency is greatly improved at a relatively low modification cost.

4.3 Parkinson's disease

In order to accurately deliver various drugs to the pathogenic site of PD, appropriate targeted delivery vectors are indispensable. Due to the particularity of PD, researchers have some unique designs in the construction of intracerebral targeting delivery vectors. At present, few homing peptides has been developed for precise delivery in the brain. In the novel treatment of PD, many therapeutic strategies aim at the recovery of mitochondrial function or the targeted elimination of ROS. Thus, the diversified modification of peptide is relatively rare.

In the research of Wang et al., exosomes derived from stem cells were used for tissue repair and nerve regeneration (Fig. 5) [251]. It also has the ability to penetrate the BBB. In order to enhance the targeting selectivity to PD microenvironment, L-arginine derivatives were selected as artificial modules binding to natural exosomes. The guanidine group can react with iNOS and ROS highly expressed in the PD microenvironment to produce nitric oxide (NO), forming a NO-driven nanomotors [252]. Meanwhile, the concentration gradient of iNOS and ROS around PD microenvironment would induce the engineered exosomes moving to microenvironment through the affinity between enzymes and substrates [253]. This dynamic design can better target the lesions of PD. Another novel strategy mainly focused on the ferroptosis in neurons. Lei et al. propose the design of a pioneering multifunctional nanoregulator, desferriamine (DFO) integrated nanosheet (BDPR NSs), which targets labile iron pool (LIP) to limit iron apoptosis and prevent PD [254]. RVG29 was introduced to target and penetrate the BBB. Similarly, ApoE peptide was fused on the protein nanocarriers based on Dps to enhance the penetration towards BBB in Zhang's work [255]. Gene drug miR-124 was delivered to the brain, reduced neuroinflammation and loss of dopaminergic neurons, and some neurobehavioral deficits were restored. In the foreseeable future, homing peptides targeting α-syn will definitely be used more widely in Parkinson's disease to promote nanoparticle bioavailability.

5 Conclusions and outlook

The incidence of CNS diseases continues to rise, with most remaining incurable. While various new drugs are being developed, the optimal delivery strategy is of paramount importance in combating these diseases. There are numerous challenges in intracerebral drug delivery, with tissue distribution being the central issue. However, due to the low permeability of the blood-brain barrier and the complexity of brain structures and partitions, the efficient and precise release of drugs remains a major challenge in delivery research. Significant progress has been made in strategies for crossing the BBB and targeting pathological areas, which vary greatly across different delivery systems. The development of peptides as flexible and adaptive ligands for carriers has shown remarkable potential for application in nanomaterials. Phage library display and analysis of various cell surface receptor structures have identified numerous peptide motifs capable of interacting with specific domains or microenvironments. Driven by innovations in computational technology for protein structure prediction and design, the three-dimensional structures of most proteins and peptides can now be accurately modeled [256]. Following the successive emergence of transformative algorithms such as Rosetta, MPNN, and RFdiffusion, the computational design of proteins or peptides tailored to bind specific antigens with high affinity and stability has become feasible [257–260]. This approach can further screen for variants amenable to soluble expression. In the near future, knowledge of the complete genome may enable the computational design of diverse novel blood-brain barrier penetrating peptides. These peptides could potentially engage currently unidentified receptors, facilitating highly efficient internalization and uptake of delivery vehicles, thereby enabling true multiplexed targeting [261]. The identification of specific receptors necessitates the application of multi-omics technologies to characterize membrane protein families, elucidating their physiological functions and pathway involvement. This represents a highly active area of research within contemporary biomedicine [262–263].

Peptide modification can enhance the BBB permeability of carriers and enable nanoparticles to be accurately targeted to specific brain regions, greatly improving tissue distribution. To our knowledge, the use of nanocarriers is a universal strategy capable of addressing the delivery challenges of multiple drugs. Peptides are important representatives of hormones in mammals, exhibiting acceptable immunogenicity and biocompatibility, while also

facilitating highly specific interactions with receptors. These advantages make peptides highly suitable for the modification of nanocarriers. Cell-penetrating peptides, BBB shuttle peptides, and homing peptides have been applied in numerous studies, demonstrating their potential in the treatment of CNS diseases. Stepwise and multidimensional targeting is a key strategy in the development of these heterogeneous carriers. When peptide modifications are combined with protein-based nanocarriers, a complete protein-based nanomaterial can be achieved. Ferritin serves as an appropriate example. The N-terminal, DE-loop, and BC-loop located on the outer surface of the ferritin 24-mer serve as natural presentation sites for peptide motifs, allowing a single ferritin subunit to carry multiple peptides. By combining this with the heterogeneity of the natural ferritin heteropolymer, complex component loading can be achieved on its surface. Although the function and significance of peptides have been thoroughly explored, their practical application remains limited. The specific mechanisms by which peptides, especially CPPs and homing peptides, achieve targeted functions remain unclear. There is also a lack of a research foundation for the precise improvement of drug delivery efficiency and tissue distribution strategies based on peptides, both in clinical studies and in cellular and animal model research. This strategy requires systematic and rigorous studies to support its application.

Data availability

Not applicable.

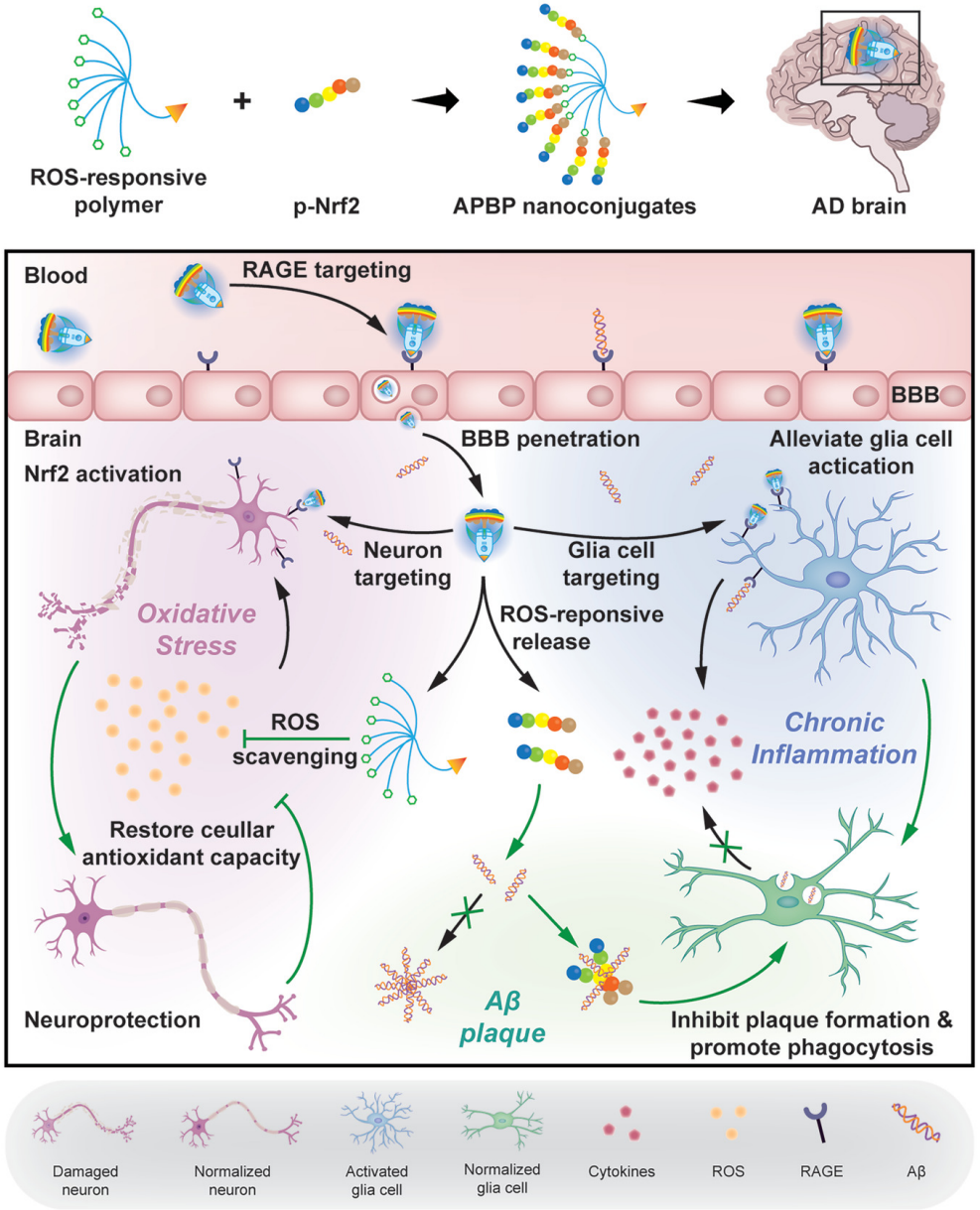

Figure 4 Illustration of APBP nanoconjugate formation and AD microenvironment modulation: (1) Brain parenchyma accumulation and damaged cells uptake via RAGE targeting; (2) ROS-responsive release of ROS scavenging polymer and Nrf2 signaling pathway activating p-Nrf2; and (3) multitarget therapy is achieved by restoring cellular antioxidant capacity, alleviating glial cell activation, and promoting Aβ phagocytosis. Reproduced with permission from Ref. [250], © Wiley-VCH GmbH 2021.

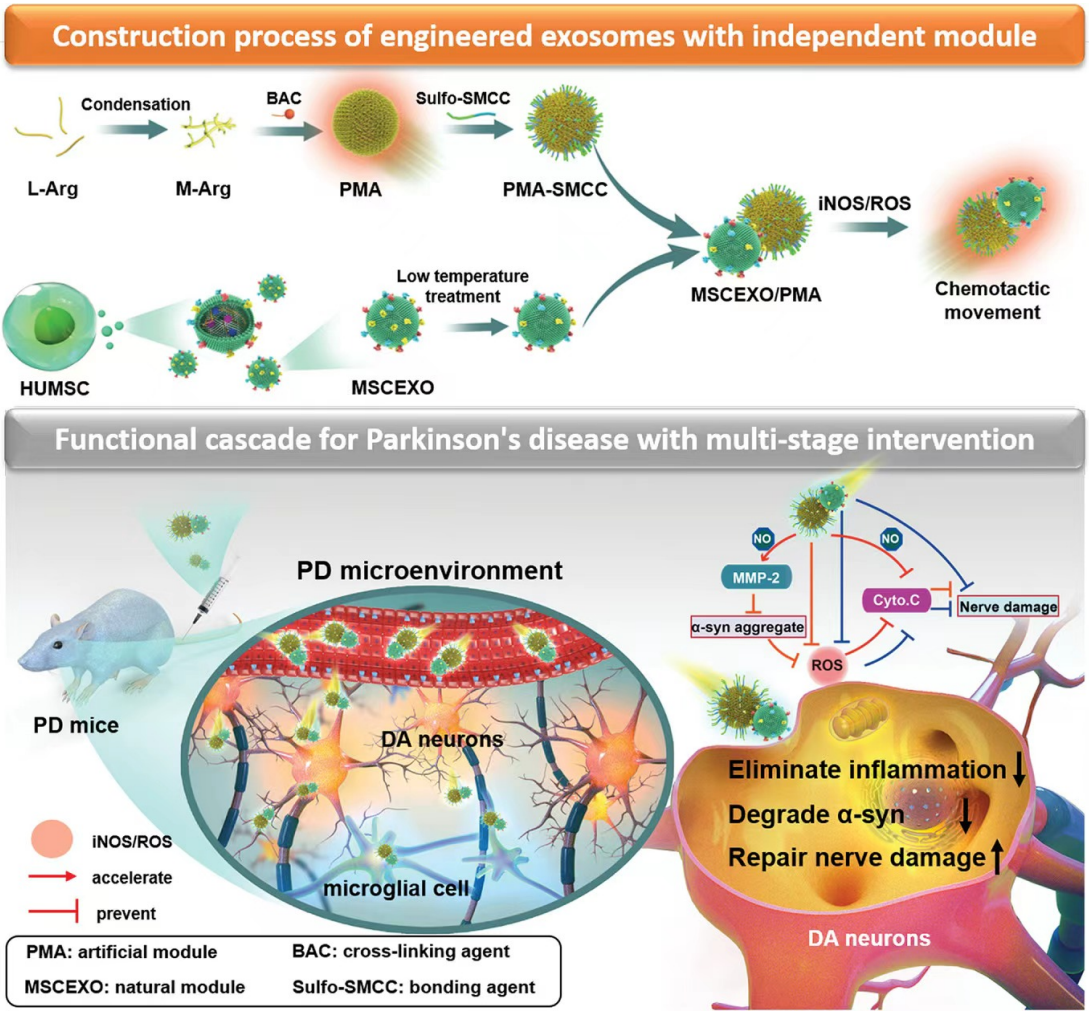

Figure 5 Schematic illustration showing the construction process of engineered exosomes with independent modules and their cascading function for the therapy of Parkinson's disease by multistep targeting and multistage intervention method (the blue line represents the action pathway of the natural exosome module MSCEXO; the red line represents the action pathway of the artificial module PMA; DA neurons represents dopaminergic neurons). Reproduced with permission from Ref. [251]. © Wiley-VCH GmbH 2022.

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Declaration of competing interest

All the contributing authors report no conflict of interests in this work.

Author contribution statement

Y. R. Q.: Writing – original draft, writing – review & editing, visualization. X. X. C.: Writing – review & editing. C. Y. L.: Conceptualization, resources. J. C. Z.: Visualization. G. H. Z.: Conceptualization, funding acquisition. T. Z.: Writing – review & editing, conceptualization, funding acquisition. All the authors have approved the final manuscript.

Informed consent

Not applicable.

Ethics statement

Not applicable.

Use of AI statement

None.

References

[1] Li, T. Q.; Zhen, H.; Wu, W. W.; Yang, F. T.; Cao, Z. H. tsRNAs: A prospective, effective therapeutic intervention for neurodegenerative diseases. *CNS Neurosci. Ther.* **2024**, *30*, e70177.

[2] Mu, R. Q.; Zhu, D. Z.; Abdulmalik, S.; Wijekoon, S.; Wei, G.; Kumbar, S. G. Stimuli-responsive peptide assemblies: Design, self-assembly, modulation, and biomedical applications. *Bioact. Mater.* **2024**, *35*, 181–207.

[3] Engelhardt, B.; Carare, R. O.; Bechmann, I.; Flügel, A.; Laman, J. D.; Weller, R. O. Vascular, glial, and lymphatic immune gateways of the central nervous system. *Acta Neuropathol.* **2016**, *132*, 317–338.

[4] Kierdorf, K.; Masuda, T.; Jordão, M. J. C.; Prinz, M. Macrophages at CNS interfaces: Ontogeny and function in health and disease. *Nat. Rev. Neurosci.* **2019**, *20*, 547–562.

[5] Prinz, M.; Priller, J. The role of peripheral immune cells in the CNS

in steady state and disease. *Nat. Neurosci.* **2017**, *20*, 136–144.
[6] Huang, L.; Huang, X. H.; Yang, X.; Hu, J. Q.; Zhu, Y. Z.; Yan, P. Y.; Xie, Y. Novel nano-drug delivery system for natural products and their application. *Pharmacol. Res.* **2024**, *201*, 107100.
[7] Qian, S.; Wang, Q. W.; Zuo, Z. Improved brain uptake of peptide-based CNS drugs via alternative routes of administrations of its nanocarrier delivery systems: A promising strategy for CNS targeting delivery of peptides. *Expert Opin. Drug Metab. Toxicol.* **2014**, *10*, 1491–1508.
[8] Kusuhara, H.; Sugiyama, Y. Efflux transport systems for drugs at the blood-brain barrier and blood-cerebrospinal fluid barrier (Part 1). *Drug Discov. Today* **2001**, *6*, 150–156.
[9] Pardridge, W. M. Recent developments in peptide drug delivery to the brain. *Pharmacol. Toxicol.* **1992**, *71*, 3–10.
[10] Abbott, N. J.; Patabendige, A. A. K.; Dolman, D. E. M.; Yusof, S. R.; Begley, D. J. Structure and function of the blood-brain barrier. *Neurobiol. Dis.* **2010**, *37*, 13–25.
[11] Daneman, R.; Prat, A. The blood-brain barrier. *Cold Spring Harbor Perspect. Biol.* **2015**, *7*, a020412.
[12] Zhao, Z.; Nelson, A. R.; Betsholtz, C.; Zlokovic, B. V. Establishment and dysfunction of the blood-brain barrier. *Cell* **2015**, *163*, 1064–1078.
[13] Nagpal, K.; Singh, S. K.; Mishra, D. N. Drug targeting to brain: A systematic approach to study the factors, parameters and approaches for prediction of permeability of drugs across BBB. *Expert Opin. Drug Deliv.* **2013**, *10*, 927–955.
[14] Oller-Salvia, B.; Sánchez-Navarro, M.; Giralt, E.; Teixidó, M. Blood-brain barrier shuttle peptides: An emerging paradigm for brain delivery. *Chem. Soc. Rev.* **2016**, *45*, 4690–4707.
[15] Redzic, Z. B.; Segal, M. B. The structure of the choroid plexus and the physiology of the choroid plexus epithelium. *Adv. Drug Deliv. Rev.* **2004**, *56*, 1695–1716.
[16] Sarin, H. Physiologic upper limits of pore size of different blood capillary types and another perspective on the dual pore theory of microvascular permeability. *J. Angiogenes. Res.* **2010**, *2*, 14.
[17] Strazielle, N.; Ghersi-Egea, J. F. Choroid plexus in the central nervous system: Biology and physiopathology. *J. Neuropathol. Exp. Neurol.* **2000**, *59*, 561–574.
[18] Saunders, N. R.; Dziegielewska, K. M.; Fame, R. M.; Lehtinen, M. K.; Liddelow, S. A. The choroid plexus: A missing link in our understanding of brain development and function. *Physiol. Rev.* **2023**, *103*, 919–956.
[19] Pellegrini, L.; Bonfio, C.; Chadwick, J.; Begum, F.; Skehel, M.; Lancaster, M. A. Human CNS barrier-forming organoids with cerebrospinal fluid production. *Science* **2020**, *369*, eaaz5626.
[20] Solár, P.; Zamani, A.; Kubíčková, L.; Dubový, P.; Joukal, M. Choroid plexus and the blood-cerebrospinal fluid barrier in disease. *Fluids Barriers CNS* **2020**, *17*, 35.
[21] Lu, Y. F.; Li, C.; Chen, Q. J.; Liu, P. X.; Guo, Q.; Zhang, Y.; Chen, X. L.; Zhang, Y. J.; Zhou, W. X.; Liang, D. H. et al. Microthrombus-targeting micelles for neurovascular remodeling and enhanced microcirculatory perfusion in acute ischemic stroke. *Adv. Mater.* **2019**, *31*, 1808361.
[22] Tuo, Q. Z.; Zhang, S. T.; Lei, P. Mechanisms of neuronal cell death in ischemic stroke and their therapeutic implications. *Med. Res. Rev.* **2022**, *42*, 259–305.
[23] Lipton, P. Ischemic cell death in brain neurons. *Physiol. Rev.* **1999**, *79*, 1431–1568.
[24] Zhu, W. B.; Libal, N. L.; Casper, A.; Bodhankar, S.; Offner, H.; Alkayed, N. J. Recombinant T cell receptor ligand treatment improves neurological outcome in the presence of tissue plasminogen activator in experimental ischemic stroke. *Transl. Stroke Res.* **2014**, *5*, 612–617.
[25] Powers, W. J.; Rabinstein, A. A.; Ackerson, T.; Adeoye, O. M.; Bambakidis, N. C.; Becker, K.; Biller, J.; Brown, M.; Demaerschalk, B. M.; Hoh, B. et al. Guidelines for the early management of patients with acute ischemic stroke: 2019 update to the 2018 guidelines for the early management of acute ischemic stroke: A guideline for healthcare professionals from the American Heart Association/American Stroke Association. *Stroke* **2019**, *50*, e344–e418.
[26] Gusev, E. I.; Martynov, M. Y.; Nikonov, A. A.; Shamalov, N. A.; Semenov, M. P.; Gerasimets, E. A.; Yarovaya, E. B.; Semenov, A. M.; Archakov, A. I.; Markin, S. S. et al. Non-immunogenic recombinant staphylokinase versus alteplase for patients with acute ischaemic stroke 4.5 h after symptom onset in Russia (FRIDA): A randomised, open label, multicentre, parallel-group, non-inferiority trial. *Lancet Neurol.* **2021**, *20*, 721–728.
[27] Song, H. Q.; Wang, Y.; Ma, Q. F.; Chen, H. S.; Liu, B.; Yang, Y.; Zhu, J. G.; Zhao, S. G.; Jin, X. P.; Li, Y. Q. et al. Efficacy and safety of recombinant human prourokinase in acute ischemic stroke: A phase IIa randomized clinical trial. *Transl. Stroke Res.* **2022**, *13*, 995–1004.
[28] Ma, G. T.; Sun, X.; Cheng, H. R.; Burgin, W. S.; Luo, W. L.; Jia, W. H.; Liu, Y. J.; He, W. L.; Geng, X. K.; Zhu, L. F. et al. Combined approach to eptifibatide and thrombectomy in acute ischemic stroke because of large vessel occlusion: A matched-control analysis. *Stroke* **2022**, *53*, 1580–1588.
[29] Barreto, A. D.; Ford, G. A.; Shen, L.; Pedroza, C.; Tyson, J.; Cai, C. Y.; Rahbar, M. H.; Grotta, J. C.; ARTSS-2 Investigators. Randomized, multicenter trial of ARTSS-2 (argatroban with recombinant tissue plasminogen activator for acute stroke). *Stroke* **2017**, *48*, 1608–1616.
[30] Chouchani, E. T.; Pell, V. R.; Gaude, E.; Aksentijević, D.; Sundier, S. Y.; Robb, E. L.; Logan, A.; Nadtochiy, S. M.; Ord, E. N. J.; Smith, A. C. et al. Ischaemic accumulation of succinate controls reperfusion injury through mitochondrial ROS. *Nature* **2014**, *515*, 431–435.
[31] Murphy, E.; Steenbergen, C. Mechanisms underlying acute protection from cardiac ischemia-reperfusion injury. *Physiol. Rev.* **2008**, *88*, 581–609.
[32] Burwell, L. S.; Nadtochiy, S. M.; Brookes, P. S. Cardioprotection by metabolic shut-down and gradual wake-up. *J. Mol. Cell. Cardiol.* **2009**, *46*, 804–810.
[33] Eltzschig, H. K.; Eckle, T. Ischemia and reperfusion-from mechanism to translation. *Nat. Med.* **2011**, *17*, 1391–1401.
[34] Zhou, Z. H.; Lu, J. F.; Liu, W. W.; Manaenko, A.; Hou, X. H.; Mei, Q. Y.; Huang, J. L.; Tang, J. P.; Zhang, J. H.; Yao, H. H. et al. Advances in stroke pharmacology. *Pharmacol. Ther.* **2018**, *191*, 23–42.
[35] Kim, J. S.; Lee, K. B.; Park, J. H.; Sung, S. M.; Oh, K.; Kim, E. G.; Chang, D. I.; Hwang, Y. H.; Lee, E. J.; Kim, W. K. et al. Safety and Efficacy of otaplimastat in patients with acute ischemic stroke requiring tPA (SAFE-TPA): A multicenter, randomized, double-blind, placebo-controlled phase 2 study. *Ann. Neurol.* **2020**, *87*, 233–245.
[36] Lyden, P.; Pryor, K. E.; Coffey, C. S.; Cudkowicz, M.; Conwit, R.; Jadhav, A.; Sawyer, R. N. Jr. ; Claassen, J.; Adeoye, O.; Song, S. et al. Final results of the RHAPSODY trial: A multi-center, phase 2 trial using a continual reassessment method to determine the safety and tolerability of 3K3A-APC, a recombinant variant of human activated protein C, in combination with tissue plasminogen activator, mechanical thrombectomy or both in moderate to severe acute ischemic stroke. *Ann. Neurol.* **2019**, *85*, 125–136.
[37] Liu, X. Y.; Yang, M.; Lei, F.; Wang, Y. R.; Yang, M. Y.; Mao, C. B. Highly effective stroke therapy enabled by genetically engineered viral nanofibers. *Adv. Mater.* **2022**, *34*, 2201210.
[38] Lo, E. H.; Dalkara, T.; Moskowitz, M. A. Mechanisms, challenges and opportunities in stroke. *Nat. Rev. Neurosci.* **2003**, *4*, 399–414.
[39] Nih, L. R.; Carmichael, S. T.; Segura, T. Hydrogels for brain repair after stroke: An emerging treatment option. *Curr. Opin. Biotechnol.* **2016**, *40*, 155–163.
[40] Carmeliet, P. Mechanisms of angiogenesis and arteriogenesis. *Nat.*

Med. **2000**, *6*, 389–395.
[41] Chen, Y. C.; Wu, J. S.; Yang, S. T.; Huang, C. Y.; Chang, C.; Sun, G. Y.; Lin, T. N. Stroke, angiogenesis and phytochemicals. *Front. Biosci. (Schol. Ed.)* **2012**, *4*, 599–610.
[42] Fang, J.; Wang, Z.; Miao, C. Y. Angiogenesis after ischemic stroke. *Acta Pharmacol. Sin.* **2023**, *44*, 1305–1321.
[43] Livingston, G.; Huntley, J.; Liu, K. Y.; Costafreda, S. G.; Selbæk, G.; Alladi, S.; Ames, D.; Banerjee, S.; Burns, A.; Brayne, C. et al. Dementia prevention, intervention, and care: 2024 report of the *Lancet* standing commission. *Lancet* **2024**, *404*, 572–628.
[44] Nandi, A.; Counts, N.; Chen, S. M.; Seligman, B.; Tortorice, D.; Vigo, D.; Bloom, D. E. Global and regional projections of the economic burden of Alzheimer's disease and related dementias from 2019 to 2050: A value of statistical life approach. *eClinicalMedicine* **2022**, *51*, 101580.
[45] Knopman, D. S.; Amieva, H.; Petersen, R. C.; Chételat, G.; Holtzman, D. M.; Hyman, B. T.; Nixon, R. A.; Jones, D. T. Alzheimer disease. *Nat. Rev. Dis. Primers* **2021**, *7*, 33.
[46] Scheltens, P.; Blennow, K.; Breteler, M. M. B.; de Strooper, B.; Frisoni, G. B.; Salloway, S.; Van der Flier, W. M. Alzheimer's disease. *Lancet* **2016**, *388*, 505–517.
[47] Ju, Y. J.; Tam, K. Y. Pathological mechanisms and therapeutic strategies for Alzheimer's disease. *Neural Regen. Res.* **2022**, *17*, 543–549.
[48] Savelieff, M. G.; Nam, G.; Kang, J.; Lee, H. J.; Lee, M.; Lim, M. H. Development of multifunctional molecules as potential therapeutic candidates for Alzheimer's disease, Parkinson's disease, and Amyotrophic Lateral Sclerosis in the last decade. *Chem. Rev.* **2019**, *119*, 1221–1322.
[49] Long, J. M.; Holtzman, D. M. Alzheimer disease: An update on pathobiology and treatment strategies. *Cell* **2019**, *179*, 312–339.
[50] Wilson, D. M.; Cookson, M. R.; Van Den Bosch, L.; Zetterberg, H.; Holtzman, D. M.; Dewachter, I. Hallmarks of neurodegenerative diseases. *Cell* **2023**, *186*, 693–714.
[51] Canter, R. G.; Penney, J.; Tsai, L. H. The road to restoring neural circuits for the treatment of Alzheimer's disease. *Nature* **2016**, *539*, 187–196.
[52] Ho, D.; Quake, S. R.; McCabe, E. R. B.; Chng, W. J.; Chow, E. K.; Ding, X. T.; Gelb, B. D.; Ginsburg, G. S.; Hassenstab, J.; Ho, C. M. et al. Enabling technologies for personalized and precision medicine. *Trends Biotechnol.* **2020**, *38*, 497–518.
[53] Ben-Shlomo, Y.; Darweesh, S.; Llibre-Guerra, J.; Marras, C.; San Luciano, M.; Tanner, C. The epidemiology of Parkinson's disease. *Lancet* **2024**, *403*, 283–292.
[54] Surmeier, D. J. Calcium, ageing, and neuronal vulnerability in Parkinson's disease. *Lancet Neurol.* **2007**, *6*, 933–938.
[55] Poewe, W.; Seppi, K.; Tanner, C. M.; Halliday, G. M.; Brundin, P.; Volkmann, J.; Schrag, A. E.; Lang, A. E. Parkinson disease. *Nat. Rev. Dis. Primers* **2017**, *3*, 17013.
[56] Spillantini, M. G.; Schmidt, M. L.; Lee, V. M. Y.; Trojanowski, J. Q.; Jakes, R.; Goedert, M. α-synuclein in Lewy bodies. *Nature* **1997**, *388*, 839–840.
[57] Bengoa-Vergniory, N.; Roberts, R. F.; Wade-Martins, R.; Alegre-Abarrategui, J. Alpha-synuclein oligomers: A new hope. *Acta Neuropathol.* **2017**, *134*, 819–838.
[58] Winner, B.; Jappelli, R.; Maji, S. K.; Desplats, P. A.; Boyer, L.; Aigner, S.; Hetzer, C.; Loher, T.; Vilar, M.; Campioni, S. et al. *In vivo* demonstration that α-synuclein oligomers are toxic. *Proc. Natl. Acad. Sci. USA* **2011**, *108*, 4194–4199.
[59] Alam, P.; Bousset, L.; Melki, R.; Otzen, D. E. α-synuclein oligomers and fibrils: A spectrum of species, a spectrum of toxicities. *J. Neurochem.* **2019**, *150*, 522–534.
[60] Lorenzen, N.; Nielsen, S. B.; Buell, A. K.; Kaspersen, J. D.; Arosio, P.; Vad, B. S.; Paslawski, W.; Christiansen, G.; Valnickova-Hansen, Z.; Andreasen, M. et al. The role of stable α-synuclein oligomers in the molecular events underlying amyloid formation. *J. Am. Chem. Soc.* **2014**, *136*, 3859–3868.
[61] Del Tredici, K.; Braak, H. Review: Sporadic Parkinson's disease: Development and distribution of α-synuclein pathology. *Neuropathol. Appl. Neurobiol.* **2016**, *42*, 33–50.
[62] Rial Verde, E. M.; Lee-Osbourne, J.; Worley, P. F.; Malinow, R.; Cline, H. T. Increased expression of the immediate-early gene *Arc/Arg3.1* reduces AMPA receptor-mediated synaptic transmission. *Neuron* **2006**, *52*, 461–474.
[63] Dijkstra, A. A.; Voorn, P.; Berendse, H. W.; Groenewegen, H. J.; Bank, N. B.; Rozemuller, A. J. M.; van de Berg, W. D. J. Stage-dependent nigral neuronal loss in incidental Lewy body and Parkinson's disease. *Mov. Disord.* **2014**, *29*, 1244–1251.
[64] Aliakbari, F.; Mohammad-Beigi, H.; Abbasi, S.; Rezaei-Ghaleh, N.; Lermyte, F.; Parsafar, S.; Becker, S.; Tafreshi, A. P.; O'Connor, P. B.; Collingwood, J. F. et al. Multiple protective roles of nanoliposome-incorporated baicalein against alpha-Synuclein aggregates. *Adv. Funct. Mater.* **2021**, *31*, 2007765.
[65] Vijiaratnam, N.; Simuni, T.; Bandmann, O.; Morris, H. R.; Foltynie, T. Progress towards therapies for disease modification in Parkinson's disease. *Lancet Neurol.* **2021**, *20*, 559–572.
[66] Paumier, K. L.; Luk, K. C.; Manfredsson, F. P.; Kanaan, N. M.; Lipton, J. W.; Collier, T. J.; Steece-Collier, K.; Kemp, C. J.; Celano, S.; Schulz, E. et al. Intrastriatal injection of pre-formed mouse α-synuclein fibrils into rats triggers α-synuclein pathology and bilateral nigrostriatal degeneration. *Neurobiol. Dis.* **2015**, *82*, 185–199.
[67] Kordower, J. H.; Chu, Y. P.; Hauser, R. A.; Freeman, T. B.; Olanow, C. W. Lewy body-like pathology in long-term embryonic nigral transplants in Parkinson's disease. *Nat. Med.* **2008**, *14*, 504–506.
[68] Chen, Z. M.; Yang, Y. P.; Yang, X.; Zhou, C. Q.; Li, F. Q.; Lei, P.; Zhong, L.; Jin, X.; Peng, G. G. Immune effects of optimized DNA vaccine and protective effects in a MPTP model of Parkinson's disease. *Neurol. Sci.* **2013**, *34*, 1559–1570.
[69] Sanchez-Guajardo, V.; Annibali, A.; Jensen, P. H.; Romero-Ramos, M. α-Synuclein vaccination prevents the accumulation of Parkinson disease-like pathologic inclusions in striatum in association with regulatory T cell recruitment in a rat model. *J. Neuropathol. Exp. Neurol.* **2013**, *72*, 624–645.
[70] Orenstein, S. J.; Kuo, S. H.; Tasset, I.; Arias, E.; Koga, H.; Fernandez-Carasa, I.; Cortes, E.; Honig, L. S.; Dauer, W.; Consiglio, A. et al. Interplay of LRRK2 with chaperone-mediated autophagy. *Nat. Neurosci.* **2013**, *16*, 394–406.
[71] Do, J.; McKinney, C.; Sharma, P.; Sidransky, E. Glucocerebrosidase and its relevance to Parkinson disease. *Mol. Neurodegener.* **2019**, *14*, 36.
[72] Chan, C. S.; Guzman, J. N.; Ilijic, E.; Mercer, J. N.; Rick, C.; Tkatch, T.; Meredith, G. E.; Surmeier, D. J. 'Rejuvenation' protects neurons in mouse models of Parkinson's disease. *Nature* **2007**, *447*, 1081–1086
[73] Ilijic, E.; Guzman, J. N.; Surmeier, D. J. The L-type channel antagonist isradipine is neuroprotective in a mouse model of Parkinson's disease. *Neurobiol. Dis.* **2011**, *43*, 364–371.
[74] Martin-Bastida, A.; Ward, R. J.; Newbould, R.; Piccini, P.; Sharp, D.; Kabba, C.; Patel, M. C.; Spino, M.; Connelly, J.; Tricta, F. et al. Brain iron chelation by deferiprone in a phase 2 randomised double-blinded placebo controlled clinical trial in Parkinson's disease. *Sci. Rep.* **2017**, *7*, 1398.
[75] Gendelman, H. E.; Zhang, Y. N.; Santamaria, P.; Olson, K. E.; Schutt, C. R.; Bhatti, D.; Shetty, B. L. D.; Lu, Y. M.; Estes, K. A.; Standaert, D. G. et al. Evaluation of the safety and immunomodulatory effects of sargramostim in a randomized, double-blind phase 1 clinical Parkinson's disease trial. *npj Parkinson's Dis.* **2017**, *3*, 10.
[76] Jucaite, A.; Svenningsson, P.; Rinne, J. O.; Cselényi, Z.; Varnäs, K.; Johnström, P.; Amini, N.; Kirjavainen, A.; Helin, S.; Minkwitz, M. et al. Effect of the myeloperoxidase inhibitor AZD3241 on microglia: A PET study in Parkinson's disease. *Brain.* **2015**, *138*,

2687–2700.
[77] Exner, N.; Lutz, A. K.; Haass, C.; Winklhofer, K. F. Mitochondrial dysfunction in Parkinson's disease: Molecular mechanisms and pathophysiological consequences. *EMBO J.* **2012**, *31*, 3038–3062.
[78] Obrador, E.; Moreno-Murciano, P.; Oriol-Caballo, M.; López-Blanch, R.; Pineda, B.; Gutiérrez-Arroyo, J. L.; Loras, A.; Gonzalez-Bonet, L. G.; Martinez-Cadenas, C.; Estrela, J. M. et al. Glioblastoma therapy: Past, present and future. *Int. J. Mol. Sci.* **2024**, *25*, 2529.
[79] Ahmed, M. H.; Canney, M.; Carpentier, A.; Thanou, M.; Idbaih, A. Unveiling the enigma of the blood-brain barrier in glioblastoma: Current advances from preclinical and clinical studies. *Curr. Opin. Oncol.* **2023**, *35*, 522–528.
[80] Hu, Y.; Li, Z. X.; Zhang, Y. C.; Wu, Y. Z.; Liu, Z. H.; Zeng, J. H.; Hao, Z. X.; Li, J.; Ren, J. Y.; Yao, M. J. The evolution of tumor microenvironment in gliomas and its implication for target therapy. *Int. J. Biol. Sci.* **2023**, *19*, 4311–4326.
[81] Schaff, L. R.; Mellinghoff, I. K. Glioblastoma and other primary brain malignancies in adults: A review. *JAMA* **2023**, *329*, 574–587.
[82] Malliou, A.; Mitsiou, C.; Kyritsis, A. P.; Alexiou, G. A. Therapeutic hypothermia in treating glioblastoma: A review. *Ther. Hypothermia Temp. Manag.* **2024**, *14*, 2–9.
[83] Yalamarty, S. S. K.; Filipczak, N.; Li, X.; Subhan, A.; Parveen, F.; Ataide, J. A.; Rajmalani, B. A.; Torchilin, V. P. Mechanisms of resistance and current treatment options for glioblastoma multiforme (GBM). *Cancers* **2023**, *15*, 2116.
[84] Eckerdt, F.; Platanias, L. C. Emerging role of glioma stem cells in mechanisms of therapy resistance. *Cancers* **2023**, *15*, 3458.
[85] Jakimovski, D.; Bittner, S.; Zivadinov, R.; Morrow, S. A.; Benedict, R. H.; Zipp, F.; Weinstock-Guttman, B. Multiple sclerosis. *Lancet* **2024**, *403*, 183–202.
[86] Bebo, B.; Cintina, I.; LaRocca, N.; Ritter, L.; Talente, B.; Hartung, D.; Ngorsuraches, S.; Wallin, M.; Yang, G. The economic burden of multiple sclerosis in the United States: Estimate of direct and indirect costs. *Neurology* **2022**, *98*, e1810–e1817.
[87] Husseini, L.; Geladaris, A.; Weber, M. S. Toward identifying key mechanisms of progression in multiple sclerosis. *Trends Neurosci.* **2024**, *47*, 58–70.
[88] Calabrese, M.; Preziosa, P.; Scalfari, A.; Colato, E.; Marastoni, D.; Absinta, M.; Battaglini, M.; De Stefano, N.; Di Filippo, M.; Hametner, S. et al. Determinants and biomarkers of progression independent of relapses in multiple sclerosis. *Ann. Neurol.* **2024**, *96*, 1–20.
[89] Cunha-Oliveira, T.; Montezinho, L.; Simões, R. F.; Carvalho, M.; Ferreiro, E.; Silva, F, S. G. Mitochondria: A promising convergent target for the treatment of amyotrophic lateral sclerosis. *Cells* **2024**, *13*, 248.
[90] Smith, E. F.; Shaw, P. J.; De Vos, K. J. The role of mitochondria in amyotrophic lateral sclerosis. *Neurosci. Lett.* **2019**, *710*, 132933.
[91] Cunha-Oliveira, T.; Montezinho, L.; Mendes, C.; Firuzi, O.; Saso, L.; Oliveira, P. J.; Silva, F. S. G. Oxidative stress in amyotrophic lateral sclerosis: Pathophysiology and opportunities for pharmacological intervention. *Oxid. Med. Cell. Longev.* **2020**, *2020*, 5021694.
[92] Feldman, E. L.; Goutman, S. A.; Petri, S.; Mazzini, L.; Savelieff, M. G.; Shaw, P. J.; Sobue, G. Amyotrophic lateral sclerosis. *Lancet* **2022**, *400*, 1363–1380.
[93] Krishna, V.; Mindel, J.; Sammartino, F.; Block, C.; Dwivedi, A. K.; Van Gompel, J. J.; Fountain, N.; Fisher, R. A phase 1 open-label trial evaluating focused ultrasound unilateral anterior thalamotomy for focal onset epilepsy. *Epilepsia* **2023**, *64*, 831–842.
[94] Jeanmonod, D.; Werner, B.; Morel, A.; Michels, L.; Zadicario, E.; Schiff, G.; Martin, E. Transcranial magnetic resonance imaging-guided focused ultrasound: Noninvasive central lateral thalamotomy for chronic neuropathic pain. *Neurosurg. Focus* **2012**, *32*, E1.
[95] Krishna, V.; Sammartino, F.; Rezai, A. A review of the current therapies, challenges, and future directions of transcranial focused ultrasound technology: Advances in diagnosis and treatment. *JAMA Neurol.* **2018**, *75*, 246–254.
[96] Song, K. H.; Harvey, B. K.; Borden, M. A. State-of-the-art of microbubble-assisted blood-brain barrier disruption. *Theranostics* **2018**, *8*, 4393–4408.
[97] McDannold, N.; Vykhodtseva, N.; Hynynen, K. Effects of acoustic parameters and ultrasound contrast agent dose on focused-ultrasound induced blood-brain barrier disruption. *Ultrasound Med. Biol.* **2008**, *34*, 930–937.
[98] Jung, O.; Thomas, A.; Burks, S. R.; Dustin, M. L.; Frank, J. A.; Ferrer, M.; Stride, E. Neuroinflammation associated with ultrasound-mediated permeabilization of the blood-brain barrier. *Trends Neurosci.* **2022**, *45*, 459–470.
[99] Kovacs, Z. I.; Kim, S.; Jikaria, N.; Qureshi, F.; Milo, B.; Lewis, B. K.; Bresler, M.; Burks, S. R.; Frank, J. A. Disrupting the blood-brain barrier by focused ultrasound induces sterile inflammation. *Proc. Natl. Acad. Sci. USA* **2017**, *114*, E75–E84.
[100] Kamimura, H. A. S.; Conti, A.; Toschi, N.; Konofagou, E. E. Ultrasound neuromodulation: Mechanisms and the potential of multimodal stimulation for neuronal function assessment. *Front. Phys.* **2020**, *8*, 150.
[101] Zhang, T. T.; Pan, N.; Wang, Y. P.; Liu, C. Y.; Hu, S. M. Transcranial focused ultrasound neuromodulation: A review of the excitatory and inhibitory effects on brain activity in human and animals. *Front. Hum. Neurosci.* **2021**, *15*, 749162.
[102] Stanimirovic, D. B.; Sandhu, J. K.; Costain, W. J. Emerging technologies for delivery of biotherapeutics and gene therapy across the blood-brain barrier. *BioDrugs* **2018**, *32*, 547–559.
[103] Mehdizadeh, S.; Mamaghani, M.; Hassanikia, S.; Pilehvar, Y.; Ertas, Y. N. Exosome-powered neuropharmaceutics: Unlocking the blood-brain barrier for next-gen therapies. *J. Nanobiotechnol.* **2025**, *23*, 329.
[104] Choi, E. S.; Shusta, E. V. Strategies to identify, engineer, and validate antibodies targeting blood-brain barrier receptor-mediated transcytosis systems for CNS drug delivery. *Expert Opin. Drug Deliv.* **2023**, *20*, 1789–1800.
[105] Liang, Y. J. et al. Engineering exosomes for targeted drug delivery. *Theranostics* **2021**, *11*, 3183–3195.
[106] Abdelsalam, M.; Ahmed, M.; Osaid, Z.; Hamoudi, R.; Harati, R. Insights into exosome transport through the blood-brain barrier and the potential therapeutical applications in brain diseases. *Pharmaceuticals* **2023**, *16*, 571.
[107] Henninot, A.; Collins, J. C.; Nuss, J. M. The current state of peptide drug discovery: Back to the Future. *J. Med. Chem.* **2018**, *61*, 1382–1414.
[108] Chow, B. W.; Gu, C. H. The molecular constituents of the blood-brain barrier. *Trends Neurosci.* **2015**, *38*, 598–608.
[109] Pardridge, W. M. Receptor-mediated peptide transport through the blood-brain barrier. *Endocr. Rev.* **1986**, *7*, 314–330.
[110] Lichota, J.; Skjørringe, T.; Thomsen, L. B.; Moos, T. Macromolecular drug transport into the brain using targeted therapy. *J. Neurochem.* **2010**, *113*, 1–13.
[111] Yu, Y. J.; Zhang, Y.; Kenrick, M.; Hoyte, K.; Luk, W.; Lu, Y. M.; Atwal, J.; Elliott, J. M.; Prabhu, S.; Watts, R. J. et al. Boosting brain uptake of a therapeutic antibody by reducing its affinity for a transcytosis target. *Sci. Transl. Med.* **2011**, *3*, 84ra44.
[112] Kreuter, J.; Hekmatara, T.; Dreis, S.; Vogel, T.; Gelperina, S.; Langer, K. Covalent attachment of apolipoprotein A-I and apolipoprotein B-100 to albumin nanoparticles enables drug transport into the brain. *J. Control. Release* **2007**, *118*, 54–58.
[113] Pan, W. H.; Kastin, A. J.; Zankel, T. C.; van Kerkhof, P.; Terasaki, T.; Bu, G. J. Efficient transfer of receptor-associated protein (RAP) across the blood-brain barrier. *J. Cell Sci.* **2004**, *117*, 5071–5078.
[114] Qian, Z. M.; Li, H. Y.; Sun, H. Z.; Ho, K. Targeted drug delivery via the transferrin receptor-mediated endocytosis pathway. *Pharmacol.*

Rev. **2002**, *54*, 561–587.
[115] Fillebeen, C.; Descamps, L.; Dehouck, M. P.; Fenart, L.; Benaïssa, M.; Spik, G.; Cecchelli, R.; Pierce, A. Receptor-mediated transcytosis of lactoferrin through the blood-brain barrier. *J. Biol. Chem.* **1999**, *274*, 7011–7017.
[116] Demeule, M.; Poirier, J.; Jodoin, J.; Bertrand, Y.; Desrosiers, R. R.; Dagenais, C.; Nguyen, T.; Lanthier, J.; Gabathuler, R.; Kennard, M. et al. High transcytosis of melanotransferrin (P97) across the blood-brain barrier. *J. Neurochem.* **2002**, *83*, 924–933.
[117] Banks, W. A.; Kastin, A. J.; Huang, W. T.; Jaspan, J. B.; Maness, L. M. Leptin enters the brain by a saturable system independent of insulin. *Peptides* **1996**, *17*, 305–311.
[118] Schwarze, S. R.; Ho, A.; Vocero-Akbani, A.; Dowdy, S. F. *In vivo* protein transduction: Delivery of a biologically active protein into the mouse. *Science* **1999**, *285*, 1569–1572.
[119] Kumar, P.; Wu, H. Q.; McBride, J. L.; Jung, K. E.; Hee Kim, M.; Davidson, B. L.; Kyung Lee, S.; Shankar, P.; Manjunath, N. Transvascular delivery of small interfering RNA to the central nervous system. *Nature* **2007**, *448*, 39–43.
[120] Werner, H. M. et al. Peptide backbone composition and protease susceptibility: Impact of modification type, position, and tandem substitution. *ChemBioChem* **2016**, *17*, 712–718.
[121] Xi, W. H.; Hansmann, U. H. E. The effect of retro-inverse D-amino acid Aβ-peptides on Aβ-fibril formation. *J. Chem. Phys.* **2019**, *150*, 095101.
[122] Miura, T.; Lee, K. J.; Katoh, T.; Suga, H. *In vitro* selection of macrocyclic $_{L}$–α/$_{D}$–α/β/γ-hybrid peptides targeting IFN–γ/IFNGR1 protein-protein interaction. *J. Am. Chem. Soc.* **2024**, *146*, 17691–17699.
[123] Imanishi, S.; Katoh, T.; Yin, Y. Z.; Yamada, M.; Kawai, M.; Suga, H. *In vitro* selection of macrocyclic $_{D}$/$_{L}$-hybrid peptides against human EGFR. *J. Am. Chem. Soc.* **2021**, *143*, 5680–5684.
[124] Ngambenjawong, C.; Pineda, J. M. B.; Pun, S. H. Engineering an affinity-enhanced peptide through optimization of cyclization chemistry. *Bioconjug. Chem.* **2016**, *27*, 2854–2862.
[125] Lee, Y.; Kim, K. M.; Nguyen, D. L.; Jannah, F.; Seong, H. J.; Kim, J. M.; Kim, Y. P. Cyclized proteins with tags as permeable and stable cargos for delivery into cells and liposomes. *Int. J. Biol. Macromol.* **2023**, *252*, 126520.
[126] Ritchey, J. L.; Filippi, L.; Ballard, D.; Pei, D. H. Bismuth-cyclized cell-penetrating peptides. *Mol. Pharm.* **2024**, *21*, 5255–5260.
[127] Brown, L.; Vidal, A. V.; Dias, A. L.; Rodrigues, T.; Sigurdardottir, A.; Journeaux, T.; O'Brien, S.; Murray, T. V.; Ravn, P.; Papworth, M. et al. Proximity-driven site-specific cyclization of phage-displayed peptides. *Nat. Commun.* **2024**, *15*, 7308.
[128] Zheng, C. Y.; Ma, C. Y.; Bai, E. Q.; Yang, K.; Xu, R. X. Transferrin and cell-penetrating peptide dual-functioned liposome for targeted drug delivery to glioma. *Int. J. Clin. Exp. Med.* **2015**, *8*, 1658–1668.
[129] Sharma, G.; Modgil, A.; Zhong, T. C.; Sun, C. W.; Singh, J. Influence of short-chain cell-penetrating peptides on transport of doxorubicin encapsulating receptor-targeted liposomes across brain endothelial barrier. *Pharm. Res.* **2014**, *31*, 1194–1209.
[130] Bechara, C.; Sagan, S. Cell-penetrating peptides: 20 years later, where do we stand. *FEBS Lett.* **2013**, *587*, 1693–1702.
[131] Nam, S. H.; Park, J.; Koo, H. Recent advances in selective and targeted drug/gene delivery systems using cell-penetrating peptides. *Arch. Pharm. Res.* **2023**, *46*, 18–34.
[132] Wu, J. M.; Roesger, S.; Jones, N.; Hu, C. M. J.; Li, S. D. Cell-penetrating peptides for transmucosal delivery of proteins. *J. Control. Release* **2024**, *366*, 864–878.
[133] Heitz, F.; Morris, M. C.; Divita, G. Twenty years of cell-penetrating peptides: From molecular mechanisms to therapeutics. *Br. J. Pharmacol.* **2009**, *157*, 195–206.
[134] Grasnick, D.; Sternberg, U.; Strandberg, E.; Wadhwani, P.; Ulrich, A. S. Irregular structure of the HIV fusion peptide in membranes demonstrated by solid-state NMR and MD simulations. *Eur. Biophys. J.* **2011**, *40*, 529–543.
[135] Morris, M. C.; Chaloin, L.; Mery, J.; Heitz, F.; Divita, G. A novel potent strategy for gene delivery using a single peptide vector as a carrier. *Nucleic Acids Res.* **1999**, *27*, 3510–3517.
[136] Varkouhi, A. K.; Scholte, M.; Storm, G.; Haisma, H. J. Endosomal escape pathways for delivery of biologicals. *J. Control. Release* **2011**, *151*, 220–228.
[137] Alipour, M.; Hosseinkhani, S.; Sheikhnejad, R.; Cheraghi, R. Nano-biomimetic carriers are implicated in mechanistic evaluation of intracellular gene delivery. *Sci. Rep.* **2017**, *7*, 41507.
[138] Li, W. J.; Nicol, F.; Szoka, F. C. Jr. GALA: A designed synthetic pH-responsive amphipathic peptide with applications in drug and gene delivery. *Adv. Drug Deliv. Rev.* **2004**, *56*, 967–985.
[139] Schutze-Redelmeier, M. P.; Gournier, H.; Garcia-Pons, F.; Moussa, M.; Joliot, A. H.; Volovitch, M.; Prochiantz, A.; Lemonnier, F. A. Introduction of exogenous antigens into the MHC class I processing and presentation pathway by Drosophila antennapedia homeodomain primes cytotoxic T cells *in vivo*. *J. Immunol.* **1996**, *157*, 650–655.
[140] Ruczynski, J.; Wierzbicki, P. M.; Kogut-Wierzbicka, M.; Mucha, P.; Siedlecka-Kroplewska, K.; Rekowski, P. Cell-penetrating peptides as a promising tool for delivery of various molecules into the cells. *Folia Histochem. Cytobiol.* **2014**, *52*, 257–269.
[141] Shi, N. Q.; Qi, X. R.; Xiang, B.; Zhang, Y. A survey on "Trojan Horse" peptides: Opportunities, issues and controlled entry to "Troy". *J. Control. Release* **2014**, *194*, 53–70.
[142] Wojciechowska, M.; Ruczynski, J.; Rekowski, P.; Alenowicz, M.; Mucha, P.; Pieszko, M.; Miszka, A.; Dobkowski, M.; Bluijssen, H. Synthesis and hybridization studies of a new CPP-PNA conjugate as a potential therapeutic agent in atherosclerosis treatment. *Protein Pept. Lett.* **2014**, *21*, 672–678.
[143] Sayers, E. J.; Cleal, K.; Eissa, N. G.; Watson, P.; Jones, A. T. Distal phenylalanine modification for enhancing cellular delivery of fluorophores, proteins and quantum dots by cell penetrating peptides. *J. Control. Release* **2014**, *195*, 55–62.
[144] Zhang, D. D.; Wang, J. X.; Xu, D. G. Cell-penetrating peptides as noninvasive transmembrane vectors for the development of novel multifunctional drug-delivery systems. *J. Control. Release* **2016**, *229*, 130–139.
[145] Wang, F. H.; Wang, Y.; Zhang, X.; Zhang, W. J.; Guo, S. R.; Jin, F. Recent progress of cell-penetrating peptides as new carriers for intracellular cargo delivery. *J. Control. Release* **2014**, *174*, 126–136.
[146] Gao, H. L.; Zhang, S.; Cao, S. J.; Yang, Z.; Pang, Z. Q.; Jiang, X. G. Angiopep-2 and activatable cell-penetrating peptide dual-functionalized nanoparticles for systemic glioma-targeting delivery. *Mol. Pharm.* **2014**, *11*, 2755–2763.
[147] Zong, T. L.; Mei, L.; Gao, H. L.; Cai, W.; Zhu, P. J.; Shi, K. R.; Chen, J. T.; Wang, Y.; Gao, F. B.; He, Q. Synergistic dual-ligand doxorubicin liposomes improve targeting and therapeutic efficacy of brain glioma in animals. *Mol. Pharm.* **2014**, *11*, 2346–2357.
[148] Youn, P.; Chen, Y. Z.; Furgeson, D. Y. A myristoylated cell-penetrating peptide bearing a transferrin receptor-targeting sequence for neuro-targeted siRNA delivery. *Mol. Pharm.* **2014**, *11*, 486–495.
[149] Khafagy, E. S.; Kamei, N.; Fujiwara, Y.; Okumura, H.; Yuasa, T.; Kato, M.; Arime, K.; Nonomura, A.; Ogino, H.; Hirano, S. et al. Systemic and brain delivery of leptin via intranasal coadministration with cell-penetrating peptides and its therapeutic potential for obesity. *J. Control. Release* **2020**, *319*, 397–406.
[150] Lim, S.; Kim, W. J.; Kim, Y. H.; Lee, S.; Koo, J. H.; Lee, J. A.; Yoon, H.; Kim, D. H.; Park, H. J.; Kim, H. M. et al. dNP2 is a blood-brain barrier-permeable peptide enabling ctCTLA-4 protein delivery to ameliorate experimental autoimmune encephalomyelitis. *Nat. Commun.* **2015**, *6*, 8244.
[151] Kang, Y. C.; Son, M.; Kang, S.; Im, S.; Piao, Y.; Lim, K. S.; Song, M. Y.; Park, K. S.; Kim, Y. H.; Pak, Y. K. Cell-penetrating artificial mitochondria-targeting peptide-conjugated metallothionein 1A alleviates mitochondrial damage in Parkinson's disease models. *Exp. Mol. Med.* **2018**, *50*, 1–13.

[152] Kim, W. J.; Kim, G. R.; Cho, H. J.; Choi, J. M. The cysteine-containing cell-penetrating peptide AP enables efficient macromolecule delivery to T cells and controls autoimmune encephalomyelitis. *Pharmaceutics* **2021**, *13*, 1134.
[153] Jana, A.; Narula, P.; Chugh, A.; Kulshreshtha, R. Efficient delivery of anti-miR-210 using Tachyplesin, a cell penetrating peptide, for glioblastoma treatment. *Int. J. Pharm.* **2019**, *572*, 118789.
[154] Silva, S.; Alves, C.; Duarte, D.; Costa, A.; Sarmento, B.; Almeida, A. J.; Gomes, P.; Vale, N. Model Amphipathic peptide coupled with tacrine to improve its antiproliferative activity. *Int. J. Mol. Sci.* **2020**, *22*, 242.
[155] Fan, S. N.; Zheng, Y. Q.; Liu, X.; Fang, W. L.; Chen, X. Y.; Liao, W.; Jing, X. N.; Lei, M.; Tao, E. X.; Ma, Q. L. et al. Curcumin-loaded PLGA–PEG nanoparticles conjugated with B6 peptide for potential use in Alzheimer's disease. *Drug Deliv.* **2018**, *25*, 1091–1102.
[156] Lakkadwala, S.; dos Santos Rodrigues, B.; Sun, C. W.; Singh, J. Biodistribution of TAT or QLPVM coupled to receptor targeted liposomes for delivery of anticancer therapeutics to brain *in vitro* and *in vivo*. *Nanomedicine: Nanotechnol. Biol. Med.* **2020**, *23*, 102112.
[157] Rusiecka, I.; Ruczyński, J.; Kozłowska, A.; Backtrog, E.; Mucha, P.; Kocić, I.; Rekowski, P. TP10-dopamine conjugate as a potential therapeutic agent in the treatment of Parkinson's disease. *Bioconjug. Chem.* **2019**, *30*, 760–774.
[158] Kim, W.; Cho, S. B.; Jung, H. Y.; Yoo, D. Y.; Oh, J. K.; Choi, G. M.; Cho, T. G.; Kim, D. W.; Hwang, I. K.; Choi, S. Y. et al. Phosphatidylethanolamine-binding protein 1 ameliorates ischemia-induced inflammation and neuronal damage in the rabbit spinal cord. *Cells* **2019**, *8*, 1370.
[159] Meng, Y.; Sun, D.; Qin, Y. Y.; Dong, X. Y.; Luo, G. Z.; Liu, Y. Cell-penetrating peptides enhance the transduction of adeno-associated virus serotype 9 in the central nervous system. *Mol. Ther. Methods Clin. Dev.* **2021**, *21*, 28–41.
[160] Liu, H.; Zhang, W.; Ma, L. N.; Fan, L. L.; Gao, F. Y.; Ni, J. M.; Wang, R. The improved blood-brain barrier permeability of endomorphin-1 using the cell-penetrating peptide synB3 with three different linkages. *Int. J. Pharm.* **2014**, *476*, 1–8.
[161] dos Santos Rodrigues, B.; Lakkadwala, S.; Kanekiyo, T.; Singh, J. Dual-modified liposome for targeted and enhanced gene delivery into mice brain. *J. Pharmacol. Exp. Ther.* **2020**, *374*, 354–365.
[162] Huang, R. Q.; Pei, Y. Y.; Jiang, C. Enhanced gene transfer into brain capillary endothelial cells using Antp-modified DNA-loaded nanoparticles. *J. Biomed. Sci.* **2007**, *14*, 595–605.
[163] Yao, Y. Z.; Wang, J.; Liu, Y.; Qu, Y.; Wang, K. K.; Zhang, Y.; Chang, Y. X.; Yang, Z.; Wan, J.; Liu, J. F. et al. Variants of the adeno-associated virus serotype 9 with enhanced penetration of the blood-brain barrier in rodents and primates. *Nat. Biomed. Eng.* **2022**, *6*, 1257–1271.
[164] Neves, V.; Aires-da-Silva, F.; Morais, M.; Gano, L.; Ribeiro, E.; Pinto, A.; Aguiar, S.; Gaspar, D.; Fernandes, C.; Correia, J. et al. Novel peptides derived from dengue virus capsid protein translocate reversibly the blood-brain barrier through a receptor-free mechanism. *ACS Chem. Biol.* **2017**, *12*, 1257–1268.
[165] Neves-Coelho, S.; Eleutério, R. P.; Enguita, F. J.; Neves, V.; Castanho, M. A. R. B. A new noncanonical anionic peptide that translocates a cellular blood-brain barrier model. *Molecules* **2017**, *22*, 1753.
[166] Frøslev, P.; Franzyk, H.; Ozgür, B.; Brodin, B.; Kristensen, M. Highly cationic cell-penetrating peptides affect the barrier integrity and facilitates mannitol permeation in a human stem cell-based blood-brain barrier model. *Eur. J. Pharm. Sci.* **2022**, *168*, 106054.
[167] Oller-Salvia, B.; Sánchez-Navarro, M.; Ciudad, S.; Guiu, M.; Arranz-Gibert, P.; Garcia, C.; Gomis, R. R.; Cecchelli, R.; García, J.; Giralt, E. et al. MiniAp-4: A venom-inspired peptidomimetic for brain delivery. *Angew. Chem., Int. Ed.* **2016**, *55*, 572–575.
[168] Arranz-Gibert, P.; Prades, R.; Guixer, B.; Guerrero, S.; Araya, E.; Ciudad, S.; Kogan, M. J.; Giralt, E.; Teixidó, M. HAI peptide and backbone analogs-validation and enhancement of biostability and bioactivity of BBB shuttles. *Sci. Rep.* **2018**, *8*, 17932.
[169] Bera, S.; Kar, R. K.; Mondal, S.; Pahan, K.; Bhunia, A. Structural elucidation of the cell-penetrating penetratin peptide in model membranes at the atomic level: Probing hydrophobic interactions in the blood-brain barrier. *Biochemistry* **2016**, *55*, 4982–4996.
[170] Zhou, X.; Smith, Q. R.; Liu, X. L. Brain penetrating peptides and peptide-drug conjugates to overcome the blood-brain barrier and target CNS diseases. *WIREs Nanomed. Nanobiotechnol.* **2021**, *13*, e1695.
[171] Sánchez-Navarro, M.; Giralt, E. Peptide shuttles for blood-brain barrier drug delivery. *Pharmaceutics* **2022**, *14*, 1874.
[172] Kawabata, H. Transferrin and transferrin receptors update. *Free Radic. Biol. Med.* **2019**, *133*, 46–54.
[173] He, J. Y.; Fan, K. L.; Yan, X. Y. Ferritin drug carrier (FDC) for tumor targeting therapy. *J. Control. Release* **2019**, *311–312*, 288–300.
[174] Wang, B.; Tang, M. P.; Yuan, Z. W.; Li, Z. Y.; Hu, B.; Bai, X.; Chu, J. X.; Xu, X. Y.; Zhang, X. Q. Targeted delivery of a STING agonist to brain tumors using bioengineered protein nanoparticles for enhanced immunotherapy. *Bioact. Mater.* **2022**, *16*, 232–248.
[175] Mojarad-Jabali, S.; Farshbaf, M.; Hemmati, S.; Sarfraz, M.; Motasadizadeh, H.; Shahbazi Mojarrad, J.; Atyabi, F.; Zakeri-Milani, P.; Valizadeh, H. Comparison of three synthetic transferrin mimetic small peptides to promote the blood-brain barrier penetration of vincristine liposomes for improved glioma targeted therapy. *Int. J. Pharm.* **2022**, *613*, 121395.
[176] Fisher, C.; Beglova, N.; Blacklow, S. C. Structure of an LDLR-RAP complex reveals a general mode for ligand recognition by lipoprotein receptors. *Mol. Cell* **2006**, *22*, 277–283.
[177] Pedersen, N. B.; Wang, S. J.; Narimatsu, Y.; Yang, Z.; Halim, A.; Schjoldager, K. T. B. G.; Madsen, T. D.; Seidah, N. G.; Bennett, E. P.; Levery, S. B. et al. Low density lipoprotein receptor class A repeats are *O*-glycosylated in linker regions. *J. Biol. Chem.* **2014**, *289*, 17312–17324.
[178] Jiang, Y.; Yang, W. J.; Zhang, J.; Meng, F. H.; Zhong, Z. Y. Protein toxin chaperoned by LRP-1-targeted virus-mimicking vesicles induces high-efficiency glioblastoma therapy *in vivo*. *Adv. Mater.* **2018**, *30*, 1800316.
[179] Ruan, H. T.; Chai, Z. L.; Shen, Q.; Chen, X. S.; Su, B. X.; Xie, C.; Zhan, C. Y.; Yao, S. Y.; Wang, H.; Zhang, M. F. et al. A novel peptide ligand RAP12 of LRP1 for glioma targeted drug delivery. *J. Control. Release* **2018**, *279*, 306–315.
[180] Savari, M. N. Fe_3O_4@Chitosan@ZIF-8@RVG29, an anti-glioma nanoplatform guided by fixed and activated by alternating magnetic field. *Sci. Rep.* **2024**, *14*, 7000.
[181] Xin, X.; Liu, W.; Zhang, Z. A.; Han, Y.; Qi, L. L.; Zhang, Y. Y.; Zhang, X. T.; Duan, H. X.; Chen, L. Q.; Jin, M. J. et al. Efficient anti-glioma therapy through the brain-targeted RVG15-modified liposomes loading paclitaxel-cholesterol complex. *Int. J. Nanomedicine* **2021**, *16*, 5755–5776.
[182] Qin, H. Z.; Jiang, Y.; Zhang, J.; Deng, C.; Zhong, Z. Y. Oncoprotein inhibitor rigosertib loaded in ApoE-targeted smart polymersomes reveals high safety and potency against human glioblastoma in mice. *Mol. Pharm.* **2019**, *16*, 3711–3719.
[183] He, P.; Xin, W.; Schulz, P.; Sierks, M. R. Bispecific antibody fragment targeting APP and inducing α-site cleavage restores neuronal health in an Alzheimer's mouse model. *Mol. Neurobiol.* **2019**, *56*, 7420–7432.
[184] Gospodarska, E. et al. Binding studies of truncated variants of the Aβ peptide to the V-domain of the RAGE receptor reveal Aβ residues responsible for binding. *Biophysica Biochimica* **2011**, *1814*, 592–609.
[185] Liu, N.; Li, M. Y.; Xie, F.; Lv, J. Q.; Gao, X. Y.; Zhang, H.; Gao, J.; Zheng, A. P. Efficacy of mimetic viral dynein binding peptide

binding nanoparticles in blood-brain barrier model. *J. Drug Deliv. Sci. Technol.* **2022**, *74*, 103523.
[186] Liu, Y.; Li, J. F.; Shao, K.; Huang, R. Q.; Ye, L. Y.; Lou, J. N.; Jiang, C. A leptin derived 30-amino-acid peptide modified pegylated poly-$_L$-lysine dendrigraft for brain targeted gene delivery. *Biomaterials* **2010**, *31*, 5246–5257.
[187] Zhang, H. Y.; van Os, W. L.; Tian, X. B.; Zu, G. Y.; Ribovski, L.; Bron, R.; Bussmann, J.; Kros, A.; Liu, Y.; Zuhorn, I. S. Development of curcumin-loaded zein nanoparticles for transport across the blood-brain barrier and inhibition of glioblastoma cell growth. *Biomater. Sci.* **2021**, *9*, 7092–7103.
[188] Qian, W. B.; Qian, M.; Wang, Y.; Huang, J. F.; Chen, J.; Ni, L. C.; Huang, Q. F.; Liu, Q. Q.; Gong, P. P.; Hou, S. Q. et al. Combination glioma therapy mediated by a dual-targeted delivery system constructed using OMCN-PEG-Pep22/DOX. *Small* **2018**, *14*, 1801905.
[189] Yeoh, Y. Q.; Amin, A.; Cuic, B.; Tomas, D.; Turner, B. J.; Shabanpoor, F. Efficient systemic CNS delivery of a therapeutic antisense oligonucleotide with a blood-brain barrier-penetrating ApoE-derived peptide. *Biomed. Pharmacother.* **2024**, *175*, 116737.
[190] Ruan, Y. T.; Xiong, Y.; Fang, W. L.; Yu, Q.; Mai, Y. R.; Cao, Z. Y.; Wang, K. X.; Lei, M.; Xu, J. X.; Liu, Y. et al. Highly sensitive Curcumin-conjugated nanotheranostic platform for detecting amyloid-beta plaques by magnetic resonance imaging and reversing cognitive deficits of Alzheimer's disease via NLRP3-inhibition. *J. Nanobiotechnol.* **2022**, *20*, 322.
[191] Gu, J. L.; Yan, C.; Yin, S.; Wu, H.; Liu, C.; Xue, A.; Lei, X.; Zhang, N.; Geng, F. Erythrocyte membrane-coated nanocarriers modified by TGN for Alzheimer's disease. *J. Control. Release* **2024**, *366*, 448–459.
[192] Birolini, G.; Valenza, M.; Ottonelli, I.; Passoni, A.; Favagrossa, M.; Duskey, J. T.; Bombaci, M.; Vandelli, M. A.; Colombo, L.; Bagnati, R. et al. Insights into kinetics, release, and behavioral effects of brain-targeted hybrid nanoparticles for cholesterol delivery in Huntington's disease. *J. Control. Release* **2021**, *330*, 587–598.
[193] Maxian, T.; Gerlitz, L.; Riedl, S.; Rinner, B.; Zweytick, D. Effect of L- to D-amino acid substitution on stability and activity of antitumor peptide RDP215 against human melanoma and glioblastoma. *Int. J. Mol. Sci.* **2021**, *22*, 8469.
[194] Peng, X. J.; Liu, X. Q.; Kim, J. Y.; Nguyen, A.; Leal, J.; Ghosh, D. Brain-penetrating peptide shuttles across the blood-brain barrier and extracellular-like space. *Bioconjug. Chem.* **2023**, *34*, 2319–2336.
[195] Majerova, P.; Hanes, J.; Olesova, D.; Sinsky, J.; Pilipcinec, E.; Kovac, A. Novel blood-brain barrier shuttle peptides discovered through the phage display method. *Molecules* **2020**, *25*, 874.
[196] Wang, Z. R.; Zhao, Y.; Hou, Y. X.; Tang, G. H.; Zhang, R. F.; Yang, Y. L.; Yan, X. Y.; Fan, K. L. A thrombin-activated peptide-templated nanozyme for remedying ischemic stroke via thrombolytic and neuroprotective actions. *Adv. Mater.* **2024**, *36*, 2210144.
[197] Hong, H. Y.; Choi, J. S.; Kim, Y. J.; Lee, H. Y.; Kwak, W.; Yoo, J.; Lee, J. T.; Kwon, T. H.; Kim, I. S.; Han, H. S. et al. Detection of apoptosis in a rat model of focal cerebral ischemia using a homing peptide selected from *in vivo* phage display. *J. Control. Release* **2008**, *131*, 167–172.
[198] Lv, W.; Xu, J. P.; Wang, X. Q.; Li, X. R.; Xu, Q. W.; Xin, H. L. Bioengineered boronic ester modified dextran polymer nanoparticles as reactive oxygen species responsive nanocarrier for Ischemic Stroke treatment. *ACS Nano* **2018**, *12*, 5417–5426.
[199] Xiao, Z. C.; Li, Y.; Chen, T.; Xie, C. C.; Liao, J.; Lin, P.; Chen, Z. S.; Qiu, Y.; Cai, J.; Zhang, C. et al. A novel nitric oxide-driven nanomotor for synergistic treatment of ischaemic stroke: Enhanced deep brain penetration and therapeutic efficacy. *Chem. Eng. J.* **2024**, *496*, 154205.
[200] Dong, Z. F.; Tang, L.; Zhang, Y.; Ma, X. Y.; Yin, Y.; Kuang, L.; Fan, Q.; Wang, B. Y.; Hu, X. Y.; Yin, T. Y. et al. A homing peptide modified neutrophil membrane biomimetic nanoparticles in response to ROS/inflammatory microenvironment for precise targeting treatment of Ischemic Stroke. *Adv. Funct. Mater.* **2024**, *34*, 2309167.
[201] Oeckl, P.; Janelidze, S.; Halbgebauer, S.; Stomrud, E.; Palmqvist, S.; Otto, M.; Hansson, O. Higher plasma β-synuclein indicates early synaptic degeneration in Alzheimer's disease. *Alzheimers Dement.* **2023**, *19*, 5095–5102.
[202] Zhang, X. C.; Zhang, X. Y.; Gao, H. L.; Qing, G. Y. Phage display derived peptides for Alzheimer's disease therapy and diagnosis. *Theranostics* **2022**, *12*, 2041–2062.
[203] Mojarad-Jabali, S.; Roh, K. H. Peptide-based inhibitors and nanoparticles: Emerging therapeutics for Alzheimer's disease. *Int. J. Pharm.* **2025**, *669*, 125055.
[204] Altendorf, T.; Gering, I.; Santiago-Schübel, B.; Aghabashlou Saisan, S.; Tamgüney, G.; Tusche, M.; Honold, D.; Schemmert, S.; Hoyer, W.; Mohrlüder, J. et al. Stabilization of monomeric tau protein by all D-enantiomeric peptide ligands as therapeutic strategy for Alzheimer's Disease and other tauopathies. *Int. J. Mol. Sci.* **2023**, *24*, 2161.
[205] Yoo, G.; Yeou, S.; Son, J. B.; Shin, Y. K.; Lee, N. K. Cooperative inhibition of SNARE-mediated vesicle fusion by α-synuclein monomers and oligomers. *Sci. Rep.* **2021**, *11*, 10955.
[206] Wei, J. L. et al. Cold beer-inspired multifunctional nanozyme for ischemic stroke with rapid thrombus clearance and long-lasting hydrogen therapy. *Nano Today* **2025**, *61*, 102636.
[207] Ayo, A.; Figueras, E.; Schachtsiek, T.; Budak, M.; Sewald, N.; Laakkonen, P. Tumor-targeting peptides: The functional screen of glioblastoma homing peptides to the target protein FABP3 (MDGI). *Cancers* **2020**, *12*, 1836.
[208] Farhangi, S.; Karimi, E.; Khajeh, K.; Hosseinkhani, S.; Javan, M. Peptide mediated targeted delivery of gold nanoparticles into the demyelination site ameliorates myelin impairment and gliosis. *Nanomedicine: Nanotechnol. Biol. Med.* **2023**, *47*, 102609.
[209] Fan, R. R.; Chuan, D.; Hou, H.; Chen, H. F.; Han, B.; Zhang, X. N.; Zhou, L. X.; Tong, A. P.; Xu, J. G.; Guo, G. Development of a hybrid nanocarrier-recognizing tumor vasculature and penetrating the BBB for glioblastoma multi-targeting therapy. *Nanoscale* **2019**, *11*, 11285–11304.
[210] Du, Y. J.; Chen, Z.; Duan, X. J.; Yan, P.; Zhang, C. L.; Kang, L.; Wang, R. F. ^{99m}Tc-labeled peptide targeting interleukin 13 receptor α 2 for tumor imaging in a cervical cancer mouse model. *Ann. Nucl. Med.* **2022**, *36*, 360–372.
[211] Yang, F. Y.; Wang, H. E.; Liu, R. S.; Teng, M. C.; Li, J. J.; Lu, M.; Wei, M. C.; Wong, T. T. Pharmacokinetic analysis of ^{111}in-labeled liposomal doxorubicin in murine glioblastoma after blood-brain barrier disruption by focused ultrasound. *PLoS One* **2012**, *7*, e45468.
[212] Charkhat Gorgich, E. A.; Kasbiyan, H.; Shabani, R.; Mehdizadeh, M.; Hajiahmadi, F.; Ajdary, M.; Barati, M.; Moradi, F.; Ahmadvand, D. Smart chlorotoxin-functionalized liposomes for sunitinib targeted delivery into glioblastoma cells. *J. Drug Deliv. Sci. Technol.* **2022**, *77*, 103908.
[213] Sevenich, M.; Honold, D.; Willuweit, A.; Kutzsche, J.; Mohrlüder, J.; Willbold, D. Development of an α-synuclein fibril and oligomer specific tracer for diagnosis of Parkinson's disease, dementia with Lewy bodies and multiple system atrophy. *Neurochem. Int.* **2022**, *161*, 105422.
[214] Özçelik, C. E.; Beğli, Ö.; Hınçer, A.; Ahan, R. E.; Kesici, M. S.; Oğuz, O.; Kasirga, T. S.; Özçubukçu, S.; Şeker, U. Ö. Ş. Synergistic screening of peptide-based biotechnological drug candidates for neurodegenerative diseases using yeast display and phage display. *ACS Chem. Neurosci.* **2023**, *14*, 3609–3621.
[215] Larbanoix, L.; Burtea, C.; Ansciaux, E.; Laurent, S.; Mahieu, I.; Vander Elst, L.; Muller, R. N. Design and evaluation of a 6-mer amyloid-beta protein derived phage display library for molecular targeting of amyloid plaques in Alzheimer's disease: Comparison with two cyclic heptapeptides derived from a randomized phage display library. *Peptides* **2011**, *32*, 1232–1243.
[216] Zhang, C. et al. Dual-functional nanoparticles targeting amyloid

plaques in the brains of Alzheimer's disease mice. *Biomaterials* **2014**, *35*, 456–465.

[217] Wang, A. T.; Huang, J. L.; Ji, M. X.; Huang, Y. K.; Chen, L.; Peng, Y. D.; Wang, C. Y.; Shi, K. X.; Zhang, C. Y.; Yu, R. H. et al. Membrane budding inspired biomimetic nanocarrier delivers brain-derived neurotrophic factor to improve AD cognition. *Adv. Funct. Mater.* **2025**, *35*, 2416572.

[218] Du, Z.; Gao, N.; Guan, Y. J.; Ding, C.; Sun, Y. H.; Ren, J. S.; Qu, X. G. Rational design of a "sense and treat" system to target amyloid aggregates related to Alzheimer's disease. *Nano Res.* **2018**, *11*, 1987–1997.

[219] Wang, F.; Zhou, X. L.; Yang, Q. G.; Xu, W. H.; Wang, F.; Chen, Y. P.; Chen, G. H. A peptide that binds specifically to the β-amyloid of Alzheimer's disease: Selection and assessment of anti-β-amyloid neurotoxic effects. *PLoS One* **2011**, *6*, e27649.

[220] Jiang, N.; Frenzel, D.; Schartmann, E.; van Groen, T.; Kadish, I.; Shah, N. J.; Langen, K. J.; Willbold, D.; Willuweit, A. Blood-brain barrier penetration of an Aβ-targeted, arginine-rich, $_{D}$-enantiomeric peptide. *Biochim. Biophys. Acta* **2016**, *1858*, 2717–2724.

[221] Saleh, S. R.; Khamiss, S. E.; Aly Madhy, S.; Khattab, S. N.; Sheta, E.; Elnozahy, F. Y.; Thabet, E. H.; Ghareeb, D. A.; Awad, D.; El-Bessoumy, A. A. Biochemical investigation and *in silico* analysis of the therapeutic efficacy of Ipriflavone through Tet-1 Surface-Modified-PLGA nanoparticles in Streptozotocin-Induced Alzheimer's like Disease: Reduced oxidative damage and etiological descriptors. *Int. J. Pharm.* **2025**, *669*, 125021.

[222] Minicozzi, V.; Chiaraluce, R.; Consalvi, V.; Giordano, C.; Narcisi, C.; Punzi, P.; Rossi, G. C.; Morante, S. Computational and experimental studies on β-sheet breakers targeting $A\beta_{1-40}$ fibrils. *J. Biol. Chem.* **2014**, *289*, 11242–11252.

[223] Zhang, Y. X.; Wang, S. W.; Lu, S.; Zhang, L. X.; Liu, D. Q.; Ji, M.; Wang, W. Y.; Liu, R. T. A mimotope of Aβ oligomers may also behave as a β-sheet inhibitor. *FEBS Lett.* **2017**, *591*, 3615–3624.

[224] Horsley, J. R.; Jovcevski, B.; Wegener, K. L.; Yu, J. X.; Pukala, T. L.; Abell, A. D. Rationally designed peptide-based inhibitor of $A\beta_{42}$ fibril formation and toxicity: A potential therapeutic strategy for Alzheimer's disease. *Biochem. J.* **2020**, *477*, 2039–2054.

[225] Zhang, X. C.; Zhang, X. Y.; Zhong, M. L.; Zhao, P.; Guo, C.; Li, Y.; Wang, T.; Gao, H. L. Selection of a $_{D}$-enantiomeric peptide specifically binding to PHF6 for inhibiting tau aggregation in transgenic mice. *ACS Chem. Neurosci.* **2020**, *11*, 4240–4253.

[226] Malhis, M.; Kaniyappan, S.; Aillaud, I.; Chandupatla, R. R.; Ramirez, L. M.; Zweckstetter, M.; Horn, A. H. C.; Mandelkow, E.; Sticht, H.; Funke, S. A. Potent tau aggregation inhibitor D-peptides selected against tau-repeat 2 using mirror image phage display. *ChemBioChem* **2021**, *22*, 3049–3059.

[227] Zhang, W. D.; Liu, Q. Y.; Haqqani, A. S.; Leclerc, S.; Liu, Z. Y.; Fauteux, F.; Baumann, E.; Delaney, C. E.; Ly, D.; Star, A. T. et al. Differential expression of receptors mediating receptor-mediated transcytosis (RMT) in brain microvessels, brain parenchyma and peripheral tissues of the mouse and the human. *Fluids Barriers CNS* **2020**, *17*, 47.

[228] Lin, J. Y.; Yu, Z. H.; Gao, X. L. Advanced noninvasive strategies for the brain delivery of therapeutic proteins and peptides. *ACS Nano* **2024**, *18*, 22752–22779.

[229] Zenaro, E.; Piacentino, G.; Constantin, G. The blood-brain barrier in Alzheimer's disease. *Neurobiol. Dis.* **2017**, *107*, 41–56.

[230] Donahue, J. E.; Flaherty, S. L.; Johanson, C. E.; Duncan III, J. A.; Silverberg, G. D.; Miller, M. C.; Tavares, R.; Yang, W. T.; Wu, Q.; Sabo, E. et al. RAGE, LRP-1, and amyloid-beta protein in Alzheimer's disease. *Acta Neuropathol.* **2006**, *112*, 405–415.

[231] Storck, S. E.; Hartz, A. M. S.; Bernard, J.; Wolf, A.; Kachlmeier, A.; Mahringer, A.; Weggen, S.; Pahnke, J.; Pietrzik, C. U. The concerted amyloid-beta clearance of LRP1 and ABCB1/P-gp across the blood-brain barrier is linked by PICALM. *Brain Behav. Immun.* **2018**, *73*, 21–33.

[232] Moloney, A. M.; Griffin, R. J.; Timmons, S.; O'Connor, R.; Ravid, R.; O'Neill, C. Defects in IGF-1 receptor, insulin receptor and IRS-1/2 in Alzheimer's disease indicate possible resistance to IGF-1 and insulin signalling. *Neurobiol. Aging* **2010**, *31*, 224–243.

[233] Lee, H.; Pienaar, I. S. Disruption of the blood-brain barrier in Parkinson's disease: Curse or route to a cure. *Front. Biosci.* **2014**, *19*, 272–280.

[234] Fujita, M.; Ichise, M.; Zoghbi, S. S.; Liow, J. S.; Ghose, S.; Vines, D. C.; Sangare, J.; Lu, J. Q.; Cropley, V. L.; Iida, H. et al. Widespread decrease of nicotinic acetylcholine receptors in Parkinson's disease. *Ann. Neurol.* **2006**, *59*, 174–177.

[235] Bartels, A. L.; Willemsen, A. T. M.; Kortekaas, R.; de Jong, B. M.; de Vries, R.; de Klerk, O.; van Oostrom, J. C. H.; Portman, A.; Leenders, K. L. Decreased blood-brain barrier P-glycoprotein function in the progression of Parkinson's disease, PSP and MSA. *J. Neural Transm.* **2008**, *115*, 1001–1009.

[236] Yuan, D. F.; Zhao, Y. L.; Banks, W. A.; Bullock, K. M.; Haney, M.; Batrakova, E.; Kabanov, A. V. Macrophage exosomes as natural nanocarriers for protein delivery to inflamed brain. *Biomaterials* **2017**, *142*, 1–12.

[237] Spiegl-Kreinecker, S.; Buchroithner, J.; Elbling, L.; Steiner, E.; Wurm, G.; Bodenteich, A.; Fischer, J.; Micksche, M.; Berger, W. Expression and functional activity of the ABC-transporter proteins P-glycoprotein and multidrug-resistance protein 1 in human brain tumor cells and astrocytes. *J. Neurooncol.* **2002**, *57*, 27–36.

[238] Choudhury, H.; Pandey, M.; Chin, P. X.; Phang, Y. L.; Cheah, J. Y.; Ooi, S. C.; Mak, K. K.; Pichika, M. R.; Kesharwani, P.; Hussain, Z. et al. Transferrin receptors-targeting nanocarriers for efficient targeted delivery and transcytosis of drugs into the brain tumors: A review of recent advancements and emerging trends. *Drug Deliv. Transl. Res.* **2018**, *8*, 1545–1563.

[239] Li, J.; Zheng, M.; Shimoni, O.; Banks, W. A.; Bush, A. I.; Gamble, J. R.; Shi, B. Y. Development of novel therapeutics targeting the blood-brain barrier: From barrier to carrier. *Adv. Sci.* **2021**, *8*, 2101090.

[240] Tiwary, S.; Morales, J. E.; Kwiatkowski, S. C.; Lang, F. F.; Rao, G.; McCarty, J. H. Metastatic brain tumors disrupt the blood-brain barrier and alter lipid metabolism by inhibiting expression of the endothelial cell fatty acid transporter Mfsd2a. *Sci. Rep.* **2018**, *8*, 8267.

[241] Geng, Z. W.; Guo, Z. L.; Guo, R. B.; Ye, R. D.; Zhu, W. S.; Yan, B. Ferroptosis and traumatic brain injury. *Brain Res. Bull.* **2021**, *172*, 212–219.

[242] Yang, Q. H.; Pu, W. D.; Hu, K. Y.; Hu, Y.; Feng, Z. Q.; Cai, J. J.; Li, C. W.; Li, L. L.; Zhou, Z. H.; Zhang, J. X. Reactive oxygen species-responsive transformable and triple-targeting butylphthalide nanotherapy for precision treatment of Ischemic Stroke by normalizing the pathological microenvironment. *ACS Nano* **2023**, *17*, 4813–4833.

[243] Zhang, H. J.; Wang, J. J.; Tian, Y. M.; Wang, C. Q.; Hou, L. Shear force-ROS sequential responsive drug delivery system for improving cerebral thrombosis microcirculation and neurological function. *J. Control. Release* **2025**, *378*, 195–208.

[244] Li, Y.; Liao, J.; Xiong, L. Y.; Xiao, Z. C.; Ye, F.; Wang, Y.; Chen, T.; Huang, L. Z.; Chen, M.; Chen, Z. S. et al. Stepwise targeted strategies for improving neurological function by inhibiting oxidative stress levels and inflammation following ischemic stroke. *J. Control. Release* **2024**, *368*, 607–622.

[245] Liu, Y.; Xia, X.; Zheng, M.; Shi, B. Y. Bio-nano toolbox for precision Alzheimer's disease gene therapy. *Adv. Mater.* **2024**, *36*, 2314354.

[246] Salloway, S.; Chalkias, S.; Barkhof, F.; Burkett, P.; Barakos, J.; Purcell, D.; Suhy, J.; Forrestal, F.; Tian, Y.; Umans, K. et al. Amyloid-related imaging abnormalities in 2 phase 3 studies evaluating aducanumab in patients with early Alzheimer Disease. *JAMA Neurol.* **2022**, *79*, 13.

[247] Wang, J.; Kong, L.; Guo, R. B.; He, S. Y.; Liu, X. Z.; Zhang, L.; Liu,

Y.; Yu, Y.; Li, X. T.; Cheng, L. Multifunctional icariin and tanshinone IIA co-delivery liposomes with potential application for Alzheimer's disease. *Drug Deliv.* **2022**, *29*, 1648–1662.

[248] Israel, L. L.; Sun, T.; Braubach, O.; Cox, A.; Shatalova, E. S.; Rashid, H. M.; Galstyan, A.; Grodzinski, Z.; Song, X. Y.; Chepurna, O. et al. β-Amyloid targeting nanodrug for neuron-specific delivery of nucleic acids in Alzheimer's disease mouse models. *J. Control. Release* **2023**, *361*, 636–658.

[249] Yang, P.; Li, Y. X.; Qian, K.; Zhou, L. L.; Cheng, Y. L.; Wu, J.; Xu, M. J.; Wang, T. Y.; Yang, X. Y.; Mu, Y. K. et al. Precise modulation of pericyte dysfunction by a multifunctional nanoprodrug to ameliorate Alzheimer's Disease. *ACS Nano* **2024**, *18*, 14348–14366.

[250] Liu, P. X.; Zhang, T. Y.; Chen, Q. J.; Li, C.; Chu, Y. C.; Guo, Q.; Zhang, Y. W.; Zhou, W. X.; Chen, H. Y.; Zhou, Z. et al. Biomimetic dendrimer-peptide conjugates for early multi-target therapy of Alzheimer's Disease by inflammatory microenvironment modulation. *Adv. Mater.* **2021**, *33*, 2100746.

[251] Wang, Q.; Li, T.; Yang, J. Y.; Zhao, Z. N.; Tan, K. Y.; Tang, S. W.; Wan, M. M.; Mao, C. Engineered exosomes with independent module/cascading function for therapy of Parkinson's Disease by multistep targeting and multistage intervention method. *Adv. Mater.* **2022**, *34*, 2201406.

[252] Wan, M. M.; Chen, H.; Wang, Q.; Niu, Q.; Xu, P.; Yu, Y. Q.; Zhu, T. Y.; Mao, C.; Shen, J. Bio-inspired nitric-oxide-driven nanomotor. *Nat. Commun.* **2019**, *10*, 966.

[253] Dey, K. K.; Das, S.; Poyton, M. F.; Sengupta, S.; Butler, P. J.; Cremer, P. S.; Sen, A. Chemotactic separation of enzymes. *ACS Nano* **2014**, *8*, 11941–11949.

[254] Lei, L.; Yuan, J. L.; Dai, Z. J.; Xiang, S.; Tu, Q. X.; Cui, X.; Zhai, S. Z.; Chen, X. Z.; He, Z. X.; Fang, B. Y. et al. Targeting the labile iron pool with engineered DFO nanosheets to inhibit ferroptosis for Parkinson's Disease therapy. *Adv. Mater.* **2024**, *36*, 2409329.

[255] Zhang, J.; Cui, B. Z.; He, T.; Hei, R. X.; Yang, L.; Liu, C.; Wu, X. N.; Wang, X.; Gao, Z. W.; Lin, F. et al. Enhancing neuroprotection in mouse model of Parkinson's Disease through protein nanosystem conjugation with ApoE peptide for miR-124 delivery. *ACS Appl. Mater. Interfaces* **2024**, *16*, 8199–8212.

[256] Abramson, J.; Adler, J.; Dunger, J.; Evans, R.; Green, T.; Pritzel, A.; Ronneberger, O.; Willmore, L.; Ballard, A. J.; Bambrick, J. et al. Accurate structure prediction of biomolecular interactions with AlphaFold 3. *Nature* **2024**, *630*, 493–500.

[257] Baek, M.; DiMaio, F.; Anishchenko, I.; Dauparas, J.; Ovchinnikov, S.; Lee, G. R.; Wang, J.; Cong, Q.; Kinch, L. N.; Schaeffer, R. D. et al. Accurate prediction of protein structures and interactions using a three-track neural network. *Science* **2021**, *373*, 871–876.

[258] Watson, J. L.; Juergens, D.; Bennett, N. R.; Trippe, B. L.; Yim, J.; Eisenach, H. E.; Ahern, W.; Borst, A. J.; Ragotte, R. J.; Milles, L. F. et al. De novo design of protein structure and function with RFdiffusion. *Nature* **2023**, *620*, 1089–1100.

[259] Dauparas, J.; Anishchenko, I.; Bennett, N.; Bai, H.; Ragotte, R. J.; Milles, L. F.; Wicky, B. I. M.; Courbet, A.; de Haas, R. J.; Bethel, N. et al. Robust deep learning-based protein sequence design using ProteinMPNN. *Science* **2022**, *378*, 49–56.

[260] Dahiyat, B. I.; Mayo, S. L. De novo protein design: Fully automated sequence selection. *Science* **1997**, *278*, 82–87.

[261] Alves, P. A.; Camargo, L. C.; Souza, G. M. D.; Mortari, M. R.; Homem-de-Mello, M. Computational modeling of pharmaceuticals with an emphasis on crossing the blood-brain barrier. *Pharmaceuticals* **2025**, *18*, 217.

[262] Sankowski, R.; Prinz, M. Human CNS-associated macrophages decoded in time and space. *Nat. Med.* **2024**, *30*, 49–50.

[263] Wälchli, T.; Bisschop, J.; Carmeliet, P.; Zadeh, G.; Monnier, P. P.; De Bock, K.; Radovanovic, I. Shaping the brain vasculature in development and disease in the single-cell era. *Nat. Rev. Neurosci.* **2023**, *24*, 271–298.