Molecular Science

AIMS Molecular Science, 11(4): 330–350.
DOI: 10.3934/molsci.2024020
Received: 01 June 2024
Revised: 13 September 2024
Accepted: 19 September 2024
Published: 27 September 2024

http://www.aimspress.com/journal/Molecular

Review

Neurogranin in Alzheimer's Disease: Roles in synaptic function, pathology, and potential as a diagnostic biomarker

Rajkumar Bavaharini[1], Chaitanya Sree Somala[2], Konda Mani Saravanan[2,3,*] and Thirunavukarasou Anand[4,*]

[1] Department of Plant Biotechnology, Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore – 641003, Tamil Nadu, India
[2] B Aatral Biosciences Private Limited, Bangalore 560091, Karnataka, India
[3] Department of Biotechnology, Bharath Institute of Higher Education and Research, Chennai 600073, Tamil Nadu, India
[4] SRIIC Lab, Faculty of Clinical Research, Sri Ramachandra Institute of Higher Education and Research, Chennai 600116, Tamil Nadu, India

* **Correspondence:** Email: saravananbioinform@bharathuniv.ac.in; anand.sriic@gmail.com.

Abstract: Postsynaptic protein neurogranin (Ng), which plays a role in synaptic plasticity, learning, and memory, has been identified as the candidate biomarker of Alzheimer's disease (AD). Cortical Amyloid β pathology seems to accelerate the onset of clinical symptoms; therefore, it is potentially valuable for early diagnosis of AD and therapeutic intervention. Synaptic pathology was shown to be an early feature of AD. Thus, proteins involved in synaptic function, such as Ng, are of great interest in studying the disease. Some prior human studies have found that Ng, a protein involved in the regulation of synaptic function, is present at greater levels in the cerebrospinal fluid of people with AD compared with those without the disease. High levels of neurogranin are associated with increased levels of synaptic vulnerability and decreased cognitive function in AD patients. This review, therefore, looked at the functionality of Ng in the brain, its association with other synaptic proteins, and its applicability as a diagnostic marker in AD. This study, therefore, sought to expand the knowledge on Ng changes in AD as it relates to synaptic dysfunction and enhanced the search for a better diagnostic and therapeutic approach.

Keywords: neurogranin; Alzheimer's disease; biomarkers; synaptic dysfunction; cognitive decline

1. Introduction

Alzheimer's disease (AD) is the most common type of dementia, impacting a large number of people globally and placing substantial strains on patients, families, and healthcare systems. According to a report from AD International, in 2023, there were more than 6.7 million people in the United States of America living with dementia [1]. This statistic highlights the pressing want for efficient methods of diagnosis and treatment. The difficulty in detecting AD stems from its subtle beginning and gradual advancement [2]. The symptoms frequently manifest subtly, which poses a challenge for early detection. Neuronal damage of considerable magnitude has often occurred when clinical indications become evident [3]. The progression of AD is schematically shown in Figure 1. Therefore, it is crucial to identify dependable biomarkers for prompt diagnosis, which is essential for improved disease management and intervention.

Historically, the diagnosis of AD has predominantly depended on identifying two characteristic proteins: Amyloid beta (Aβ) and tau [4, 5]. Aβ plaques and neurofibrillary tangles, which consist of tau proteins that have undergone excessive phosphorylation, are distinct characteristics identified in the brains of individuals with AD [6, 7]. These biomarkers are essential, but they have limitations in their sensitivity and specificity, especially in the first phases of the disease. Consequently, there is a burgeoning interest in investigating supplementary biomarkers that can more precisely and promptly indicate the underlying pathophysiological alterations. A promising area of study centers on biomarkers associated with synaptic dysfunction, which occurs before the noticeable loss of neurons seen in AD [8]. Synaptic dysfunction is crucial as it has a direct correlation with cognitive decline, which is the primary manifestation of AD [9]. Studying biomarkers associated with synaptic health might provide a valuable understanding of initial illness processes and prospective targets for treatment.

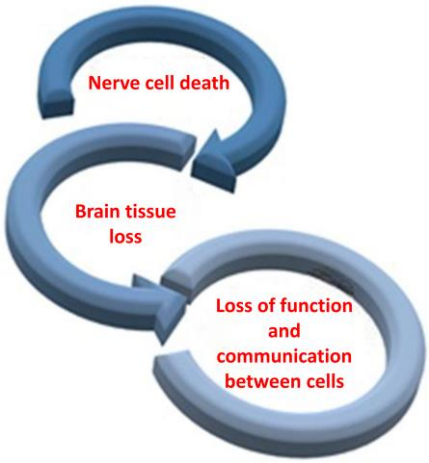

Figure 1. Progression of Alzheimer's disease.

Neurogranin (Ng) is identified as an essential biomarker in this situation [10]. Ng is a protein found mainly in the brain in areas related to memory and learning, such as the hippocampus and cortex [11]. Synaptic plasticity is crucial for learning and memory as it determines the ability of synapses to either develop or diminish over time. Furthermore, neurogranin has a role in synapse

regeneration and long-term potentiation (LTP), both essential for preserving synaptic integrity and cognitive function [12]. The interest in Ng as a biomarker for AD stems from its functions and the early changes in its expression levels in response to synaptic injury. Research has demonstrated that reduced levels of Ng in cerebrospinal fluid (CSF) are associated with the loss of synapses and impairment in cognitive function [13]. This suggests that Ng could be a valuable marker for detecting early synaptic dysfunction in AD. Moreover, alterations in Ng levels have been observed before substantial neuronal degeneration, emphasizing its potential value in early detection [14].

This review explores the significance of Ng, investigates its functions, and assesses its importance as a biomarker in AD. By comprehending the function of Ng in synaptic processes and its modifications in Alzheimer's pathology, we can acquire a more profound comprehension of the initial occurrences of the illness. This insight could facilitate the development of more precise diagnostic tools and successful therapy strategies that specifically target the synaptic components of AD. The subsequent sections will thoroughly examine Ng, elucidating its biological functions, the mechanisms that underlie its participation in synaptic function, and the evidence that supports its potential as a biomarker for AD.

2. Early detection and treatment of AD

Alzheimer's disease (AD) is a degenerative neurological condition that causes a steady decline in brain neurons, resulting in noticeable declines in memory, cognition, and behavior. Discovered by Dr. Alois Alzheimer, a German psychiatrist, in the early 20th century, this disease is characterized by unique pathological alterations in the brain. Dr. Alzheimer observed the accumulation of Aβ plaques outside of neurons, which interfere with the communication between neurons, and the formation of neurofibrillary tangles made up of tau proteins inside neurons, which block the transport of necessary nutrients [15,16]. The presence of these anomalies emphasizes the intricate neuropathology that underlies AD and emphasizes the difficulties in detecting and treating this incapacitating disorder [17].

A variety of variables, including genetics, environment, and lifestyle choices, cause Alzheimer's disease. Notable unchangeable risk factors comprise older age, genetics, and family history, greatly enhancing an individual's vulnerability to getting the condition [1]. Although it is impossible to change these risk factors, studies have revealed certain factors that can be modified to prevent or delay AD's beginning (Figure 2). These techniques include improving cognitive reserve, such as participating in consistent mental and physical activities, fostering social relationships, and mitigating cardiovascular health issues to minimize neuropathological harm [18].

Even with these preventive efforts, the diagnosis of AD typically takes place around 2.8 years after the first signs of symptoms, resulting in delays in receiving treatment and assistance [19]. An essential difficulty in identifying AD is its similarity to other neurodegenerative illnesses, which can make the clinical presentation more complex and hide the underlying pathology [20]. Hence, an urgent need is for more accurate and early diagnostic instruments to detect AD in its initial phases. The investigation of biomarkers has arisen as a crucial field of study to tackle these diagnostic difficulties. Biomarkers are biological markers that can offer objective measurements of the existence and advancement of a disease [21]. The biomarkers aβ and tau proteins have been thoroughly investigated in AD and are essential to the current diagnostic criteria. Nevertheless, these indicators predominantly indicate subsequent clinical alterations and may not accurately represent the first phases of the disease.

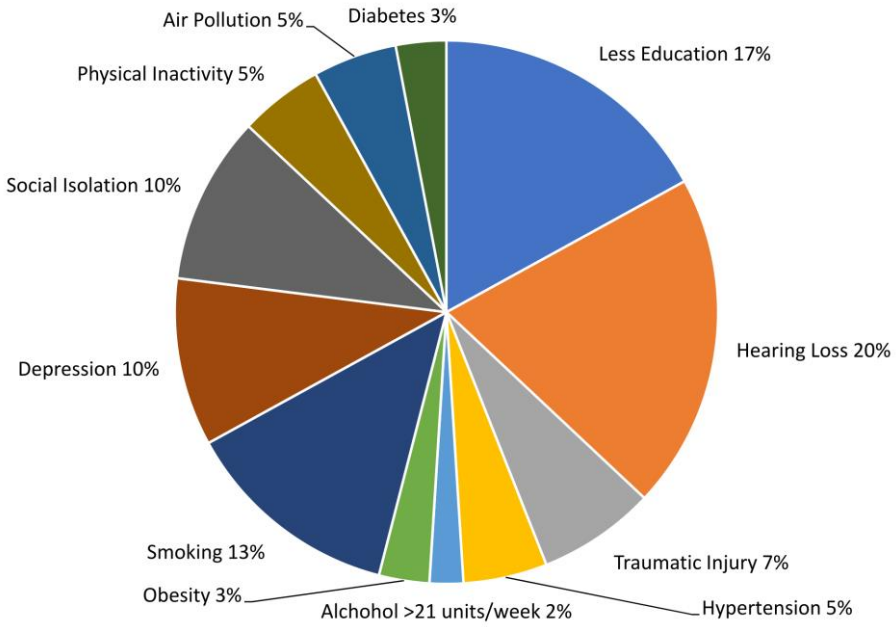

Figure 2. Modifiable risk factors of Alzheimer's disease and the percentage of reduction in the prevalence if eliminated.

Current studies have focused on examining biomarkers linked to synaptic disruption, which occurs at an early stage of the disease and is strongly connected to the reduction in cognitive abilities [22]. Ng is an essential biomarker for synaptic plasticity, synaptic regeneration, and long-term potentiation. It is a postsynaptic protein. Studies have demonstrated a decline in Ng levels in the cerebrospinal fluid (CSF) of persons diagnosed with Alzheimer's disease [23]. This decrease is associated with the loss of synapses and cognitive decline. Neurogranin shows excellent potential as a candidate for early detection and targeted treatment of AD [24]. An efficient way of evaluating neurodegeneration in AD is by establishing synaptic degeneration by the levels of synaptic proteins in CSF [25]. Neuronal loss has been accepted for a long time as one of the main hallmarks of the neurodegenerative process and is regarded as the best predictor of cognitive impairment in AD [26]. Of the multiple synaptic proteins, neurogranin is the most promising fluid biomarker of synaptic injury [25]. We have also discovered that increased levels of neurogranin in the CSF were present in AD patients and that it was directly proportional to the patient's deterioration in cognitive function [27,28]. For example, a correlation has been established between Ng and hippocampal volume and the brain metabolic rate assessed by fluorodeoxyglucose positron emission tomography (FDG PET), pointing to the significance of Ng as the marker of synaptic and cognitive integrity [29]. Further, CSF-Ng has been an outcome measure in clinical trials for lecanemab, a monoclonal antibody against amyloid beta, demonstrating its uses in evaluating treatment response [30]. In addition to neurogranin, other synaptic proteins, including neuromodulin (GAP43) and beta synuclein (SNCB), have also been proven to be significantly linked to AD. These proteins are also shown to be upregulated in CSF and are related to the pathophysiology of AD, including tau proteins [31].

In particular, GAP43 and SNCB were significantly associated with CSF total tau (t-tau) and phosphorylated tau (p-tau) levels, indicating that tau changes are tightly connected to synaptic dysfunction [32]. This relationship confirms the dependency between synaptic dysfunction and tau

pathology in the development of AD and suggests that synaptic protein levels should be used to evaluate neurodegeneration [33]. First, the constant co-localization of these synaptic proteins with tau pathology underscores their utility in the framework of AD pathogenesis and illustrates the possibility of utilizing these biomarkers for better comprehending disease processes and monitoring AD progression at the early phase [34].

Therefore, the assessment of synaptic proteins such as neurogranin, GAP43, and beta synuclein in CSF is not just a theoretical exercise but a practical method with significant implications for the evaluation of synaptic dysfunction and neurodegeneration in AD [35,36]. The strong associations between these proteins and conventional indices of AD pathology provide solid support for their potential as biomarkers, paving the way for the development of more targeted and effective diagnostic and therapeutic approaches that focus on preserving synaptic viability and efficiency. The following sections will provide a more detailed analysis of the biology of Ng, its role in synaptic function, and the evidence that supports its usefulness as a biomarker for AD.

3. Biomarkers in AD

One of the main obstacles in managing AD is the early and precise diagnosis of the disease, which is necessary for effective intervention and therapy. Biomarkers, defined by the Alzheimer's Association as observable biological alterations indicative of disease presence or risk, play a crucial role. By enabling the diagnosis of AD at its early stages, biomarkers can significantly improve patient outcomes. The National Institute on Aging–Alzheimer's Association (NIA-AA) has devised a comprehensive framework for identifying biomarkers in AD, known as the ATN system (Table 1). This paradigm categorizes biomarkers into three primary groups: A (Amyloid beta), T (Tau), and N (Neurodegeneration). Aβ biomarkers detect the buildup of amyloid plaques, which are extracellular deposits that impede neuronal communication. Tau biomarkers evaluate the existence of neurofibrillary tangles within neurons, which hinder nutrition transport and contribute to cell death. Neurodegeneration (N) biomarkers reveal general neuronal injury and loss, representing the amount of brain degeneration [37–39].

Table 1. ATN classification of biomarkers in Alzheimer's disease with examples.

ATN Classification	Example
Amyloid beta	Aβ38, Aβ40**,** Aβ42
Tau	tau-p181, tau-p217**,** tau-p231, tau-total
Neurodegeneration	Neurogranin, Neurofilament light polypeptide **(**NFL)

In the preclinical stage of Alzheimer's disease, pathological changes can already be detected by particular biomarkers even before the onset of clinical symptoms. This early discovery is critical as it allows timely treatment intervention, possibly reducing disease development [40]. The introduction of in vivo biomarkers, which can be tested in living individuals by non-invasive techniques, has transformed the diagnosis of AD. These biomarkers include imaging techniques such as positron emission tomography (PET) scans, which can identify amyloid plaques and tau tangles in the brain, as well as fluid biomarkers found in CSF and blood [21].

Among the diagnostic biomarkers for AD, Aβ and Tau have been extensively investigated and verified. Aβ biomarkers include Aβ42 and the ratio of Aβ42 to Aβ40, both of which are generally

evaluated in CSF. Decreased levels of Aβ42 in CSF suggest the presence of amyloid plaque buildup in the brain. Tau biomarkers, such as t-tau and p-tau, indicate the existence of neurofibrillary tangles and neuronal damage when their levels are increased in CSF [41,42].

Aside from the conventional biomarkers, numerous newly identified biomarkers are being recognized for their potential to detect and track AD at an early stage. These comprise CSF biomarkers, namely chitinase-3-like protein 1 (YKL-40), Ng, synaptosomal-associated protein-25 (SNAP-25), and visinin-like protein 1 (VILIP-1). YKL-40 is a biomarker of inflammation, and increased concentrations in CSF are linked to the presence of neuroinflammation in individuals with AD [43]. Ng is a protein found in the postsynaptic region of neurons that plays a role in the ability of synapses to change and adapt, known as synaptic plasticity. When the levels of Ng in the CSF are lower, it is associated with a decrease in synapses and a drop in cognitive function. SNAP-25 and VILIP-1 are linked to synaptic function and neuronal integrity, respectively, further understanding AD's synaptic and neuronal alterations [44].

In addition, developing new blood and genetic biomarkers improves the precision and practicality of diagnosing AD. Blood biomarkers, such as the ratio of plasma Aβ42/40, plasma p-tau, and neurofilament light chain (NfL), provide a less intrusive and more readily available method for early detection. These biomarkers have demonstrated the potential to represent the pathological changes happening in the brain accurately and have a strong correlation with CSF biomarkers and imaging results [45]. Genetic biomarkers, such as apolipoprotein E (APOE) genotyping, offer insights into the genetic risk factors linked to AD. The APOE ε4 allele is a widely recognized risk factor for AD and can be used to identify individuals at a greater risk of getting the disease [46].

Ultimately, biomarkers are essential instruments in the identification and treatment of AD. The ATN classification system offers a systematic method for classifying these biomarkers, enabling a thorough comprehension of the disease's pathogenesis. Incorporating conventional and novel biomarkers, such as CSF, blood, and genetic markers, has significant potential in enhancing the precision and promptness of diagnosing AD. Continuous research and progress in biomarker technologies are crucial for developing more efficient diagnostic tools and therapeutic approaches, ultimately improving patient care and results in AD.

4. Neurogranin: A key protein in synaptic function and AD

Neurogranin is a gene that encodes a protein found on the 11th chromosome of humans. It covers a distance of around 12.5 kilobase pairs (kbp) [39]. Ng is a gene that produces a postsynaptic protein called p17. This protein was discovered as a substrate of protein kinase C when isolated from the bovine brain [10, 47]. The term "neurogranin" is derived from the granular appearance that may be noticed under an electron microscope in the pyramidal cells of the hippocampus and cortex in the central nervous system (CNS) [47, 48]. Ng is a compact protein, weighing around 7.5 kilodaltons (kDa) and made up of 78 amino acids. The Ng gene consists of four exons and three introns in its structure. Exon 1 is responsible for encoding the initial five amino acids of the protein, whereas exon 2 is responsible for encoding the remaining 73 amino acids. Exons 3 and 4 comprise untranslated regions that are involved in the control of gene expression [10]. The protein's evolutionary conservation is highlighted by a significant level of sequence similarity between human and rat Ng, with a 90% correspondence at the nucleic acid level and a 96% correspondence at the protein level. The significant resemblance indicates that Ng plays crucial functional roles in various species [49].

Ng is mainly found in the brain, specifically in the hippocampus and cortex, and is essential for cognitive processes, including learning and remembering. This particular protein located in the postsynaptic region plays a crucial role in various essential processes, such as synaptic plasticity, synaptic regeneration, and LTP [50]. Synaptic plasticity is the capacity of synapses to undergo strengthening or weakening over a period, which is a primary mechanism that underlies the processes of learning and memory. Ng has a critical function in this process by regulating the presence of calmodulin (CaM). This messenger protein binds to calcium and is necessary for activating calcium/CaM-dependent protein kinase II (CaMKII). CaMKII is important for synaptic strength and plasticity [51].

Studies have emphasized the importance of Ng in AD. Synaptic dysfunction and loss in AD are initial occurrences that exhibit a high correlation with cognitive decline. Levels of Ng in CSF have been observed to decline in persons with AD, indicating a decrease in synaptic function and loss. Ng can potentially be a biomarker for early detection and the tracking of illness advancement [52]. In addition, alterations in Ng expression have been linked to the extent of cognitive decline, providing additional evidence for its potential usefulness in clinical environments [28]. The neuronal localization of Ng, which is observable using electron microscopy, has yielded valuable information regarding its distribution and role throughout the brain. The acceptable level of detail indicates that it is explicitly positioned in dendritic spines, tiny projections on dendrites where synapses are found. Dendritic spines are structures that exhibit morphological alterations in response to synaptic activity, and the presence of Ng in these spines implies its involvement in regulating their plasticity and stability [53].

Ng expression is regulated by neuronal activity and can be affected by factors including calcium influx and activation of protein kinase C. Furthermore, research has demonstrated that epigenetic mechanisms, including DNA methylation and histone modifications, influence the expression of Ng. This emphasizes the crucial role of Ng in preserving synaptic function and plasticity [54]. The evolutionary conservation of Ng further highlights its functional significance. The significant resemblance between the Ng sequences in humans and rats indicates that this protein has been conserved across time because of its crucial functions in synaptic activity. This conservation also enables the utilization of animal models to investigate the functions of Ng and its role in AD [55]. As more investigation delves into the intricacies of Ng's functions and regulatory mechanisms, it is anticipated to assume a progressively significant part in our comprehension of synapse biology and neurodegenerative illnesses.

5. Expression of neurogranin

Neurogranin levels of expression vary significantly in various areas of the brain and during different stages of development. This diversity indicates its essential function in developing and upkeeping neural synapses, which are vital for cognitive functions such as acquiring knowledge and retaining information [56]. During the early stages of postnatal development, the brain experiences significant growth and restructuring of its synaptic connections. The expression of Ng is especially prominent during this period, coinciding with the active phase of synapse formation [57]. The expression of Ng at different locations is listed in Table 2. Studies have shown that the levels of Ng reach their highest point from 2 to 3 years of age. This is an essential time for the formation of synapses and the ability of synapses to change, which are crucial processes for cognitive

development and functioning [22,58]. The elevated concentrations of Ng seen during this initial phase indicate its role in establishing and fine-tuning synaptic connections. During development, the expression of Ng becomes increasingly specialized in certain regions [59]. Ng is primarily located in dendritic spines, the tiny protrusions on dendrites where synapses are generated. Dendritic spines are highly adaptable structures that have a critical function in the transmission and flexibility of synapses. The presence of Ng in these spines highlights its significance in regulating synaptic strength and plasticity. The protein's capacity to attach to CaM, a crucial modulator of calcium signaling in neurons, further emphasizes its involvement in synaptic activity. Ng controls the availability of calmodulin, affecting CaMKII's activity. This activity is crucial for synaptic plasticity and LTP [60].

Table 2. Expression of neurogranin at different locations.

Study	Tracing Techniques	Expression of Neurogranin (Ng)
Svirsky et al., 2019 [61]	Immunohistochemistry	Traumatic brain injury affects neurogranin by characterizing protein expression changes at various time points after the injury.
Agnello et al., 2021 [8]	Commercial ELISA kit	Found in neurons of the cerebral cortex, hippocampus, amygdala, and striatum; also detected in certain non-neuronal tissues such as the adrenal gland and kidney.
Alberto et al., 2019 [62]	Immunofluorescence	The expression of Ng in cultured hippocampal neurons showed that protein and mRNA levels were about 10% of that found in the adult hippocampus.
Willemse et al., 2020 [29]	Fluorodeoxyglucose positron emission tomography	CSF Ng is a general biomarker for synaptic degeneration, strongly correlating with CSF tau.
Hwang et al., 2021 [63]	Immunoblotting	Altered Ng levels affect the phosphorylation landscape of neuronal proteins.
Cheriyan et al., 2020 [64]	Immunofluorescence	Human and mouse endothelia

During maturity, the level of Ng expression remains consistent in brain areas linked to advanced cognitive abilities, such as the hippocampus and cortex. These regions play a crucial role in memory formation, spatial navigation, and executive processes. The continuous presence of Ng in these regions indicates its continued function in preserving synaptic well-being and adaptability, which are crucial for cognitive resilience and adjustment to novel encounters [65]. Ng expression is regulated through an intricate mechanism that is impacted by multiple factors, such as neuronal activity and epigenetic changes. Neuronal activity, namely the entry of calcium through N-methyl-D-aspartate (NMDA) receptors, can increase the expression of Ng, hence strengthening its function in regulating synaptic plasticity. Furthermore, the regulation of Ng gene expression is also influenced by epigenetic mechanisms, including DNA methylation and histone modifications. This highlights the complex control of this essential protein [54].

Ng expression changes are particularly intriguing when considering AD. Research has indicated that persons with AD experience a drop in Ng levels in their CSF, which is a result of synapse loss

and dysfunction. Since synaptic degradation occurs early in AD, the decrease in Ng levels can be used as a biomarker to diagnose and track the evolution of the disease [52]. Additionally, alterations in Ng expression have been linked to the extent of cognitive decline in patients with AD, indicating that Ng serves as an indicator of synaptic pathology and is also connected with clinical outcomes [28]. The importance of Ng goes beyond its function in the regular operation of the brain to its potential as a target for therapeutic interventions. Modulating the levels or function of Ng could provide novel treatment options for cognitive deficiencies related to neurodegenerative illnesses due to its role in calcium signaling and synaptic plasticity. For example, increasing the level of Ng or imitating its role could enhance synaptic connections' strength and cognitive performance in situations when the integrity of synapses is damaged [66]. An ongoing study into the intricacies of Ng regulation and function shows potential for enhancing our comprehension of brain development, synaptic plasticity, and the mechanisms behind neurodegenerative diseases.

6. Functions of neurogranin

Neurogranin plays a crucial role in synaptic plasticity, mainly through mechanisms like LTP and long-term depression (LTD). These mechanisms are responsible for the cellular foundations of cognitive functions. This has been supported by studies conducted by Cardozo et al. [67], Camporesi et al. [22], and Agnello et al. [8]. Synaptic plasticity refers to the ability of synapses, the connections between neurons, to change and adapt in response to activity and experience. Synaptic plasticity pertains to the capacity of synapses to enhance or diminish in strength over time in response to changes in their activity levels [68]. LTP and LTD are two primary types of synaptic plasticity that correspond to the enhancement and reduction of synaptic connections, respectively (Figure 3). Ng plays a crucial role in these processes by interacting with CaM, an essential calcium-bound protein [69].

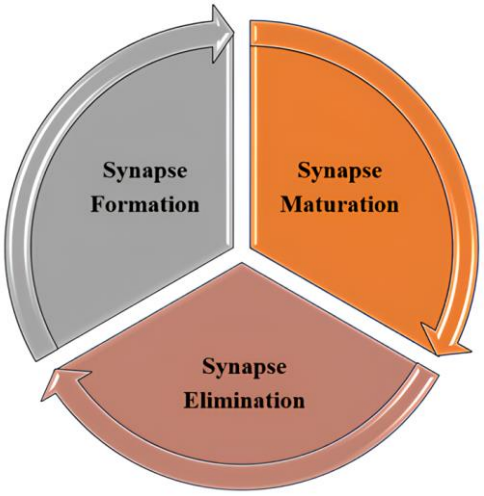

Figure 3. Series of synaptic plasticity.

LTP is a persistent increase in the transmission of signals between two neurons, which occurs when they are activated simultaneously. It is a fundamental process that forms the basis of learning and memory. As a protein that binds to CaM, Ng plays a crucial part in this process. During synaptic activity, calcium ions (Ca^{2+}) entering the postsynaptic neuron via NMDARs bind to CaM, which

subsequently activates CaMKII [68]. CaMKII, when activated, phosphorylates the α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors (AMPARs), resulting in an augmentation of synaptic transmission through an increase in the quantity of AMPARs present at the synapse and their ability to conduct signals [54,70].

Ng interacts with calmodulin when it is not phosphorylated and moves it close to the plasma membrane. The strategic placement of calmodulin guarantees its accessibility for interaction with calcium ions that arrive through NMDARs, hence expediting the swift activation of CaMKII. Ng undergoes phosphorylation in response to an increase in intracellular calcium levels. This phosphorylation leads to the release of calmodulin, which then activates CaMKII effectively. As a result, this process promotes LTP over LTD [54,70]. LTD refers to the activity-dependent weakening of neuronal synapses that lasts for hours or longer. This is in contrast to LTP. The process entails the stimulation of calcineurin (CaN). This enzyme removes phosphate groups from different synaptic proteins, resulting in the uptake of AMPARs and a consequent reduction in synaptic transmission strength. CaM activates CaN instead of CaMKII when calcium levels are reduced. Ng influences this pathway by its ability to bind to CaM. In settings of low calcium, Ng, which is not phosphorylated, binds to calmodulin, blocking its activation of CaMKII and instead promoting the activation of CaN, hence increasing LTD [54,70]. The protein's function in calcium signaling pathways facilitates synaptic plasticity and contributes to the reorganization and development of new synaptic connections (Figure 4).

Experimental studies have emphasized the functional importance of Ng in synaptic plasticity. For instance, animals that do not have Ng show significantly decreased LTP and poor memory that relies on the hippocampus. This demonstrates the crucial function of the protein in these biological processes [48,54,71]. These findings emphasize the vital role of Ng in facilitating the synaptic pathways responsible for learning and memory.

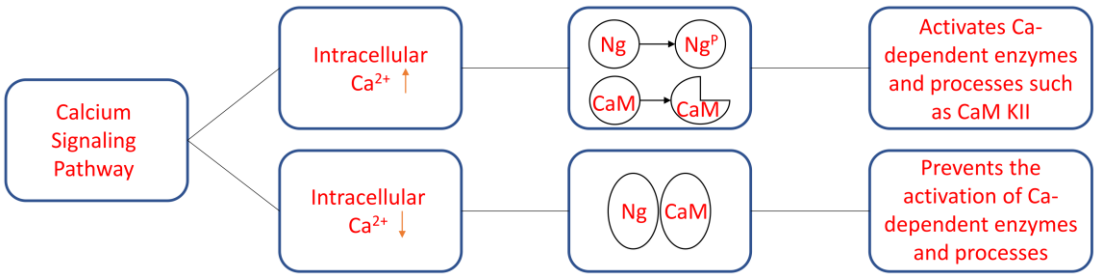

Figure 4. Diagrammatic representation of Ng in the calcium signaling pathway. An increase in intracellular calcium leads to phosphorylation of Ng and conformational changes in CaM, leading to the activation of Ca-dependent enzymes and processes, whereas if the intracellular calcium is low, then the unphosphorylated Ng binds to CaM and prevents the activation of Ca-dependent enzymes and processes.

Ng has a crucial role in controlling synaptic plasticity and regeneration. Its binding to CaM and its proximity to the plasma membrane makes it a critical regulator of calcium signaling pathways that play a role in LTP and LTD [72]. Ng exerts a substantial influence on the activation of CaMKII and CaN, hence playing a crucial role in the facilitation and impairment of synaptic connections. Ng and neuromodulin (Nm) are neuron-specific substrates of protein kinase C (PKC) that modulate the

activity of calmodulin (CaM) and are implicated in learning and memory processes [73]. Ng and Nm are intrinsically disordered proteins; however, they adopt helical conformations upon binding to CaM (Figure 5A). The structure of IQ peptides (24 amino acids) from the Nm/Ng complex with CaM reveals that key binding residues in Ng are situated in a negatively charged pocket in the apo form of CaM [74]. The phosphorylation of these residues by PKC neutralizes the primary charge of these residues and introduces a negative charge in the region, which repels Nm/Ng from CaM.

Consequently, this prevents the activation of various CaM-dependent functions related to learning and memory. This phosphorylation mechanism elucidates how PKC-induced modifications inhibit the binding of Nm/Ng to CaM, highlighting the critical role of phosphorylation in regulating synaptic signals [74]. This binding mode was demonstrated through functional analysis, revealing that an Ng mutant, in which arginine was replaced by alanine, failed to enhance synaptic transmission in hippocampal neurons. This finding indicates the importance of this residue for CaM-modulated synaptic activity. Ng binds to calmodulin in receptor binding competition, competing directly with other CaM-binding proteins, such as CaMKII [68]. Thus, competitive binding is essential for regulating synaptic plasticity mechanisms, including LTP LTD, in neurons associated with learning and memory. The competition between neurogranin and other proteins for CaM binding represents a significant regulatory mechanism in neuronal signaling pathways.

Figure 5B presents a schematic protein-protein interaction map centered on CaM and its associated proteins constructed by the String database [75]. It demonstrates the complex interactions among various members of the CaM family and neuron-specific proteins involved in synaptic signaling and function. This network comprises multiple isoforms of calmodulin, including CALM1, CALM2, CALM3, CALML3, CALML4, and CALML5, which are intricately interconnected due to their roles in calcium signaling and the regulation of subsequent events. Ng is predominantly expressed with CaM, highlighting its significance in controlling synaptic plasticity through CaM [76]. Additional proteins in the network, such as GAP43 and IQCJ, demonstrate significant binding affinities with calmodulin, highlighting their involvement in synaptic activity and plasticity [77]. The intricate interrelations among proteins in the network, such as those between CaM isoforms and kinase subunits (e.g., PHKA1, PHKB, and PHKG2), indicate a highly interconnected signaling platform centered on CaM, which seems to govern various cellular processes related to neurotransmitter release and synaptic plasticity [78]. Furthermore, CaM's interactions with synaptic proteins like SNCA (alpha-synuclein) and NEFL (neurofilament light polypeptide) highlight the significance of CaM signaling in neurodegenerative illnesses, where the disruption of these pathways may result in pathological circumstances [79]. The network map illustrates the synaptic and signaling proteins interacting with CaM, elucidating the intricate molecular interactions fundamental to neuronal function. It also highlights potential nodes of interest, particularly in the context of neurodegenerative disorders pertinent to the progression of Alzheimer's disease and other similar conditions. The connections illustrated herein emphasize the significance of CaM as the master switch molecule in the nervous system, regulating complex signaling pathways essential for synaptic homeostasis.

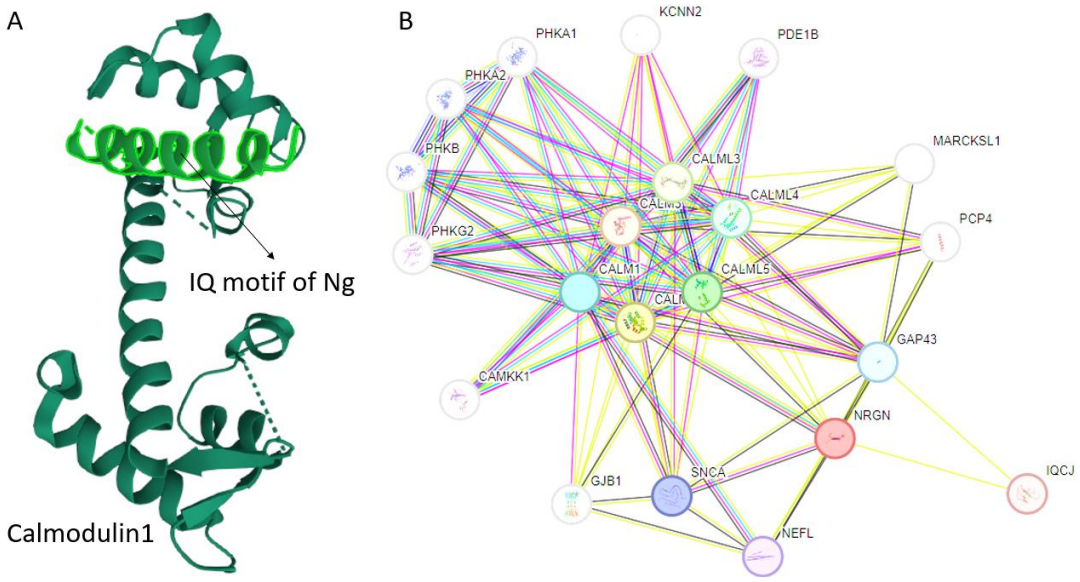

Figure 5. (A). The Protein Data Bank (PDB) identifier for the complex of IQ peptides of Ng with CaM is 4E50. This structure shows the interaction between Ng and CaM in its calcium-free (apo) state, highlighting the binding mechanism relevant to its role in synaptic plasticity and signaling. (B). This network map provides a comprehensive visualization of how CaM interacts with multiple synaptic and signaling proteins, illustrating the intricate molecular landscape that underpins neuronal function.

7. Neurogranin and AD

A comprehensive understanding of the biomarkers linked to synaptic alterations is essential for the timely identification and intervention of AD. Synaptic dysfunction is an early pathological characteristic of AD, which happens before the noticeable loss of neurons or the onset of clinical symptoms [80]. The early breakdown of synaptic connections highlights the significance of discovering biomarkers that can accurately indicate these alterations. Ng is highly valuable for this purpose because of its crucial involvement in synaptic plasticity. Studies have demonstrated that Ng levels exhibit substantial variation in several brain regions and body fluids in individuals with AD [81]. Ng levels are reduced in the brains of persons with AD, specifically in the hippocampus and cerebral cortex. These brain regions play a significant role in cognitive processes, including learning and memory, and are greatly affected by AD [48,82,83]. The decrease in Ng levels in these crucial regions indicates the decline in synaptic integrity and function. In contrast, levels of Ng in the CSF are elevated in AD. The observed rise is most likely caused by the release of Ng into the CSF due to synaptic damage and degeneration. Individuals with AD have higher levels of Ng in their CSF compared to those with mild cognitive impairment (MCI). This indicates that Ng in CSF can be used as a sensitive marker to track the progression of synaptic pathology from early to advanced stages of the disease [84].

Ng is also linked to forming Aβ and tau proteins, which are the characteristic protein buildups in AD. Aβ plaques and neurofibrillary tangles, which consist of tau proteins that have been excessively phosphorylated, interfere with synaptic function and lead to the death of neurons [85]. The participation of Ng in various pathways highlights its significance as a biomarker. Research has

demonstrated that the levels of Ng are changed in exosomes produced from neurons and separated from blood plasma [23]. Neuronal-derived extracellular vesicles (NDEs) are tiny structures secreted by neurons and contain chemical substances that mirror the characteristics of the neurons they come from. Ng levels in individuals with AD are reduced in neuronal-derived extracellular vesicles (NDEs), but there is no notable alteration in the overall levels of Ng in the blood plasma [52]. These findings indicate that changes unique to neurons in Ng can be identified without intrusive procedures, providing a possible method for early detection and tracking of AD.

Clinicians may detect AD at an earlier stage by assessing Ng levels in CSF and neurodegenerative exosomes before significant cognitive impairment occurs. Timely intervention and management of the condition are essential. Ng levels can be used to monitor the advancement of synaptic degeneration in AD [86]. This can help evaluate the effectiveness of therapeutic measures designed to maintain synaptic function. Ng can be used to distinguish AD from other neurodegenerative disorders that do not show the same Ng changes. Ng is intimately associated with synaptic abnormalities unique to AD [87]. Additional investigation is required to completely clarify the mechanisms that underlie Ng's function in AD and its interaction with Aβ and tau [88]. Furthermore, extensive clinical trials are necessary to confirm the efficacy of Ng as a dependable biomarker for diagnosing and monitoring AD [44]. Ng is a crucial protein indicating the synaptic disruption typical of AD when its expression is changed. The presence of the disease in CSF and NDEs gives vital information about the initial phases of the disease. It is a promising method for diagnosing and monitoring treatment progress [89]. As our comprehension of Ng function in AD becomes more profound, it has the potential to enhance patient outcomes by enabling earlier detection and more precise treatment techniques.

Elevated concentrations of Ng may render it beneficial in clinical contexts such as AD and other neurodegenerative disorders due to its role in synaptic plasticity and memory via the modulation of CaM [72]. Given that Ng is exclusive to neurons and synapses, elevated levels of Ng in CSF are observed in cases of synaptic degeneration and cognitive decline, facilitating early illness diagnosis and assessment of severity [90]. Furthermore, activating Ng pathways may be advantageous for treating the disorders above, as it enhances synaptic and cognitive functions. Nonetheless, the utilization of Ng as a biomarker has certain limitations that necessitate additional validation: first, the specificity and sensitivity of Ng in Alzheimer's patients; second, elevated Ng levels may also occur in other neurodegenerative disorders. Additional problems encompass diversity among measuring methods, assay conditions, and patient groups, complicating the procedure; thus, establishing standardized protocols is essential. Moreover, while Ng is recognized for its role in synaptic function, a more comprehensive understanding of the specific mechanisms of Ng targeting and potential adverse effects associated with therapeutic interventions targeting Ng is necessary, given its interactions with various CaM-binding proteins [91]. These difficulties necessitate thoroughly considering risks and returns or gains and losses. Nonetheless, Ng possesses significant potential for clinical applications, and further research will be essential to establish its utility as both a diagnostic instrument and a therapeutic target in neurodegenerative disorders.

8. Future directions

Detailed investigations on Ng should focus on elucidating the particular mechanisms implicated in synaptopathy and the role of Ng in AD. Although elevated levels of Ng in CSF are associated with

synapse loss and cognitive decline in AD patients, the precise mechanisms via which Ng influences these processes remain inadequately understood. Future research elucidating the interactions between Ng and other synaptic proteins and their impact on signaling cascades may enhance our comprehension of Ng's role in developing synaptic plasticity and associated diseases. Furthermore, analyses utilizing alternative methodologies, including advanced imaging technologies and biochemical techniques, may assist in delineating the specific spatial and temporal patterns of Ng concerning the development of amyloid β and tau pathology, thereby enhancing comprehension of the correlation between Ng alterations and various stages of AD. Another crucial area for future research is the standardization of methodologies for evaluating Ng levels in CSF and, potentially, blood. This is particularly important as Ng could serve as a biomarker for the early diagnosis of AD. Current measurement methodologies often lack accuracy, making it necessary to improve them to align more closely with clinical practice. Furthermore, future investigations could explore the incorporation of Ng as a marker alongside other indicators, such as tau and Aβ, to develop a multiparameter biomarker model. Such a model would more accurately represent the complex nature of AD pathophysiology and provide a more precise delineation of the onset and progression of the condition.

Additional research should focus on the efficacy of regulating Ng to restore synaptic function in AD. Consequently, comprehending the implications of altering Ng activity through pharmacological or gene therapies may yield novel avenues for addressing synaptic loss and cognitive deterioration in individuals with AD. Investigating the role of Ng phosphorylation and its capacity to bind CaM and other signaling molecules in the regulation of synaptic plasticity may uncover further targets that might be modified to reverse pathology. Moreover, examining the effects of Ng-targeted therapeutics in animal models and clinical trials could provide critical insights into the efficacy and dangers of these interventions, hence advancing the research of these medicines toward practical use.

9. Conclusions

Alzheimer's disease ranks as the sixth most common cause of death in individuals over the age of 65. A significant limitation in the development of therapeutic options is the inadequate diagnosis of the disease, which is attributed to its intricate etiology. Each individual in this condition presents unique symptoms. Since AD is multifactorial, investigating several biomarkers is essential for its identification. Ng is a synaptic protein that plays a role in LTP, a mechanism involved in the plasticity of synapses. Recent studies have demonstrated a link between elevated Ng levels in the CSF of individuals with AD and the presence of Aβ and tau proteins. This suggests that Ng could serve as a promising biomarker for diagnosing AD. The utilization of Ng and other biomarkers plays a significant role in the timely detection and treatment of AD, making its clinical implications crucial. Further research on the biomarkers associated with various phases of AD can aid in improving disease diagnosis and advancing novel treatments.

Author contributions

KMS and TA designed the study; RB performed data collection and literature analysis; CSS prepared figures and tables; KMS and TA prepared initial draft; KMS contributed to review and editing. All authors approved the final version of the manuscript.

Acknowledgments

The Institutional Ethics Committee of Bharath Institute of Higher Education and Research approved all of the research protocols through approval number BIHER/BT/0524/2024.

Conflict of interest

The authors declare no conflicts of interest.

References

1. Alzheimer's Association (2023) 2023 Alzheimer's disease facts and figures. *Alzheimers Dement* 19: 1598–1695. https://doi.org/10.1002/alz.13016
2. Diogo VS, Ferreira HA, Prata D, et al. (2022) Early diagnosis of Alzheimer's disease using machine learning: A multi-diagnostic, generalizable approach. *Alz Res Therapy* 14: 107. https://doi.org/10.1186/s13195-022-01047-y
3. Jakob-Roetne R, Jacobsen H (2009) Alzheimer's disease: From pathology to therapeutic approaches. *Angew Chem Int Ed Engl* 48: 3030–3059. https://doi.org/10.1002/anie.200802808
4. Ferrari-Souza JP, Bellaver B, Ferreira PCL, et al. (2023) *APOEε4* potentiates amyloid β effects on longitudinal tau pathology. *Nat Aging* 3: 1210–1218. https://doi.org/10.1038/s43587-023-00490-2
5. Saravanan KM, Kannan M, Meera P, et al. (2022) E3 ligases: A potential multi-drug target for different types of cancers and neurological disorders. *Future Med Chem* 14: 187–201. https://doi.org/10.4155/fmc-2021-0157
6. Moore KBE, Hung TJ, Fortin JS (2023) Hyperphosphorylated tau (p-tau) and drug discovery in the context of Alzheimer's disease and related tauopathies. *Drug Discov Today* 28: 103487. https://doi.org/10.1016/j.drudis.2023.103487
7. Saravanan KM, Zhang H, Zhang H, et al. (2020) On the conformational dynamics of β-amyloid forming peptides: A computational perspective. *Front Bioeng Biotechnol* 8: 532. https://doi.org/10.3389/fbioe.2020.00532
8. Agnello L, Lo Sasso B, Vidali M, et al. (2021) Neurogranin as a reliable biomarker for synaptic dysfunction in Alzheimer's disease. *Diagnostics* 11: 2339. https://doi.org/10.3390/diagnostics11122339
9. Pelucchi S, Gardoni F, Di Luca M, et al. (2022) Chapter 28 - Synaptic dysfunction in early phases of Alzheimer's Disease, In: *Handbook of Clinical Neurology*, Elsevier, 184: 417–438. https://doi.org/10.1016/B978-0-12-819410-2.00022-9
10. Saunders T, Gunn C, Blennow K, et al. (2023) Neurogranin in Alzheimer's disease and ageing: A human post-mortem study. *Neurobiol Dis* 177: 105991. https://doi.org/10.1016/j.nbd.2023.105991
11. Rani S, Dhar SB, Khajuria A, et al. (2023) Advanced overview of biomarkers and techniques for early diagnosis of Alzheimer's disease. *Cell Mol Neurobiol* 43: 2491–2523. https://doi.org/10.1007/s10571-023-01330-y
12. Abraham WC, Jones OD, Glanzman DL (2019) Is plasticity of synapses the mechanism of long-term memory storage? *npj Sci Learn* 4: 9. https://doi.org/10.1038/s41539-019-0048-y

13. Xue M, Sun FR, Ou YN, et al. (2020) Association of cerebrospinal fluid neurogranin levels with cognition and neurodegeneration in Alzheimer's disease. *Aging* 12: 9365–9379. https://doi.org/10.18632/aging.103211
14. Toader C, Dobrin N, Brehar FM, et al. (2023) From recognition to remedy: The significance of biomarkers in neurodegenerative disease pathology. *Int J Mol Sci* 24: 16119. https://doi.org/10.3390/ijms242216119
15. Scheltens P, De Strooper B, Kivipelto M, et al. (2021) Alzheimer's disease. *Lancet* 397: 1577–1590. https://doi.org/10.1016/S0140-6736(20)32205-4
16. Cardoso BR, Roberts BR, Malpas CB, et al. (2019) Supranutritional sodium selenate supplementation delivers selenium to the central nervous system: Results from a randomized controlled pilot trial in Alzheimer's disease. *Neurotherapeutics* 16: 192–202. https://doi.org/10.1007/s13311-018-0662-z
17. DeTure MA, Dickson DW (2019) The neuropathological diagnosis of Alzheimer's disease. *Mol Neurodegener* 14: 32. https://doi.org/10.1186/s13024-019-0333-5
18. Zhang XX, Tian Y, Wang ZT, et al. (2021) The Epidemiology of Alzheimer's disease modifiable risk factors and prevention. *J Prev Alzheimers Dis* 8: 313–321. https://doi.org/10.14283/jpad.2021.15
19. Dubois B, von Arnim CAF, Burnie N, et al. (2023) Biomarkers in Alzheimer's disease: Role in early and differential diagnosis and recognition of atypical variants. *Alz Res Therapy* 15: 175. https://doi.org/10.1186/s13195-023-01314-6
20. Jellinger KA (2022) Recent update on the heterogeneity of the Alzheimer's disease spectrum. *J Neural Transm* 129: 1–24. https://doi.org/10.1007/s00702-021-02449-2
21. Dubois B, Villain N, Frisoni GB, et al. (2021) Clinical diagnosis of Alzheimer's disease: Recommendations of the International Working Group. *Lancet Neurol* 20: 484–496. https://doi.org/10.1016/S1474-4422(21)00066-1
22. Camporesi E, Nilsson J, Brinkmalm A, et al. (2020) Fluid biomarkers for synaptic dysfunction and loss. *Biomark Insights* 15: 1177271920950319. https://doi.org/10.1177/1177271920950319
23. Liu W, Lin H, He X, et al. (2020) Neurogranin as a cognitive biomarker in cerebrospinal fluid and blood exosomes for Alzheimer's disease and mild cognitive impairment. *Transl Psychiatry* 10: 125. https://doi.org/10.1038/s41398-020-0801-2
24. Hindley N, Sanchez Avila A, Henstridge C (2023) Bringing synapses into focus: Recent advances in synaptic imaging and mass-spectrometry for studying synaptopathy. *Front Synaptic Neurosci* 15: 1130198. https://doi.org/10.3389/fnsyn.2023.1130198
25. Mravinacová S, Alanko V, Bergström S, et al. (2024) CSF protein ratios with enhanced potential to reflect Alzheimer's disease pathology and neurodegeneration. *Mol Neurodegener* 19: 15. https://doi.org/10.1186/s13024-024-00705-z
26. Milos T, Vuic B, Balic N, et al. (2024) Cerebrospinal fluid in the differential diagnosis of Alzheimer's disease: An update of the literature. *Expert Rev Neurother* 1–17. https://doi.org/10.1080/14737175.2024.2400683
27. Pereira JB, Janelidze S, Ossenkoppele R, et al. (2021) Untangling the association of amyloid-β and tau with synaptic and axonal loss in Alzheimer's disease. *Brain* 144: 310–324. https://doi.org/10.1093/brain/awaa395

28. Kester MI, Teunissen CE, Crimmins DL, et al. (2015) Neurogranin as a cerebrospinal fluid biomarker for synaptic loss in symptomatic Alzheimer disease. *JAMA Neurol* 72: 1275–1280. https://doi.org/10.1001/jamaneurol.2015.1867
29. Willemse EAJ, Sieben A, Somers C, et al. (2021) Neurogranin as biomarker in CSF is non-specific to Alzheimer's disease dementia. *Neurobiol Aging* 108: 99–109. https://doi.org/10.1016/j.neurobiolaging.2021.08.002
30. van Dyck CH, Swanson CJ, Aisen P, et al. (2023) Lecanemab in early Alzheimer's disease. *N Engl J Med* 388: 9–21. https://doi.org/10.1056/NEJMoa2212948
31. Bergström S, Remnestål J, Yousef J, et al. (2021) Multi-cohort profiling reveals elevated CSF levels of brain-enriched proteins in Alzheimer's disease. *Ann Clin Transl Neurol* 8: 1456–1470. https://doi.org/10.1002/acn3.51402
32. Remnestål J, Just D, Mitsios N, et al. (2016) CSF profiling of the human brain enriched proteome reveals associations of neuromodulin and neurogranin to Alzheimer's disease. *Proteomics Clin Appl* 10: 1242–1253. https://doi.org/10.1002/prca.201500150
33. Sandelius Å, Portelius E, Källén Å, et al. (2019) Elevated CSF GAP-43 is Alzheimer's disease specific and associated with tau and amyloid pathology. *Alzheimers Dement* 15: 55–64. https://doi.org/10.1016/j.jalz.2018.08.006
34. McGrowder DA, Miller F, Vaz K, et al. (2021) Cerebrospinal fluid biomarkers of Alzheimer's disease: Current evidence and future perspectives. *Brain Sci* 11: 215. https://doi.org/10.3390/brainsci11020215
35. Remnestål J, Bergström S, Olofsson J, et al. (2021) Association of CSF proteins with tau and amyloid β levels in asymptomatic 70-year-olds. *Alz Res Therapy* 13: 54. https://doi.org/10.1186/s13195-021-00789-5
36. Rosenberg A, Öhlund-Wistbacka U, Hall A, et al. (2022) β-amyloid, tau, neurodegeneration classification and eligibility for anti-amyloid treatment in a memory clinic population. *Neurology* 99: e2102–e2113. https://doi.org/10.1212/WNL.0000000000201043
37. Cummings J (2019) The national institute on aging—Alzheimer's association framework on Alzheimer's disease: Application to clinical trials. *Alzheimers Dement* 15: 172–178. https://doi.org/10.1016/j.jalz.2018.05.006
38. Tahami Monfared AA, Byrnes MJ, White LA, et al. (2022) Alzheimer's disease: Epidemiology and clinical progression. *Neurol Ther* 11: 553–569. https://doi.org/10.1007/s40120-022-00338-8
39. Xiang Y, Xin J, Le W, et al. (2020) Neurogranin: A potential biomarker of neurological and mental diseases. *Front Aging Neurosci* 12: 584743. https://doi.org/10.3389/fnagi.2020.584743
40. van der Flier WM, de Vugt ME, Smets EMA, et al. (2023) Towards a future where Alzheimer's disease pathology is stopped before the onset of dementia. *Nat Aging* 3: 494–505. https://doi.org/10.1038/s43587-023-00404-2
41. Rasmussen J, Langerman H (2019) Alzheimer's disease–why we need early diagnosis. *Degener Neurol Neuromuscul Dis* 123–130. https://doi.org/10.2147/DNND.S228939
42. Liang Y, Kang X, Zhang H, et al. (2023) Knockdown and inhibition of hippocampal GPR17 attenuates lipopolysaccharide-induced cognitive impairment in mice. *J Neuroinflammation* 20: https://doi.org/271. 10.1186/s12974-023-02958-9
43. Schindler SE, Bollinger JG, Ovod V, et al. (2019) High-precision plasma β-amyloid 42/40 predicts current and future brain amyloidosis. *Neurology* 93: e1647–e1659. https://doi.org/10.1212/WNL.0000000000008081

44. Klyucherev TO, Olszewski P, Shalimova AA, et al. (2022) Advances in the development of new biomarkers for Alzheimer's disease. *Transl Neurodegener* 11: 25. https://doi.org/10.1186/s40035-022-00296-z
45. Janelidze S, Mattsson N, Palmqvist S, et al. (2020) Plasma P-tau181 in Alzheimer's disease: Relationship to other biomarkers, differential diagnosis, neuropathology and longitudinal progression to Alzheimer's dementia. *Nat Med* 26: 379–386. https://doi.org/10.1038/s41591-020-0755-1
46. O'Bryant SE, Petersen M, Hall J, et al. (2022) Characterization of mild cognitive impairment and dementia among community-dwelling Mexican Americans and non-hispanic whites. *J Alzheimers Dis* 90: 905–915. https://doi.org/10.3233/JAD-220300
47. Pohlan J, Kress W, Hermann K-G, et al. (2020) Computed tomography thermography for ablation zone prediction in microwave ablation and cryoablation: Advantages and challenges in an ex vivo porcine liver model. *J Comput Assist Tomogr* 44: 744–749. https://doi.org/10.1097/RCT.0000000000001081
48. Agnello L, Gambino CM, Lo Sasso B, et al. (2021) Neurogranin as a novel biomarker in Alzheimer's disease. *Lab Med* 52: 188–196. https://doi.org/10.1093/labmed/lmaa062
49. Nashine S, Kenney MC (2024) Effects of Humanin G (HNG) on angiogenesis and neurodegeneration markers in Age-related Macular Degeneration (AMD). *Mitochondrion* 74: 101818. https://doi.org/10.1016/j.mito.2023.11.001
50. Vontell RT, Gober R, Dallmeier J, et al. (2024) Association of region-specific hippocampal reduction of neurogranin with inflammasome proteins in post mortem brains of Alzheimer's disease. *Alzheimers Dement* 10: e12444. https://doi.org/10.1002/trc2.12444
51. Li L, Lai M, Cole S, et al. (2020) Neurogranin stimulates Ca^{2+}/calmodulin-dependent kinase II by suppressing calcineurin activity at specific calcium spike frequencies. *PLoS Comput Biol* 16: e1006991. https://doi.org/10.1371/journal.pcbi.1006991
52. Kvartsberg H, Lashley T, Murray CE, et al. (2019) The intact postsynaptic protein neurogranin is reduced in brain tissue from patients with familial and sporadic Alzheimer's disease. *Acta Neuropathol* 137: 89–102. https://doi.org/10.1007/s00401-018-1910-3
53. Svirsky SE, Henchir J, Li Y, et al. (2024) Temporal-specific sex and injury-dependent changes on neurogranin-associated synaptic signaling after controlled cortical impact in rats. *Mol Neurobiol* 61: 7256–7268. https://doi.org/10.1007/s12035-024-04043-5
54. Zhong L, Gerges NZ (2020) Neurogranin regulates metaplasticity. *Front Mol Neurosci* 12: 322. https://doi.org/10.3389/fnmol.2019.00322
55. Rueda-García V, Rondón-Barragán IS (2024) Molecular characterization of neurogranin (NRGN) gene from Red-Bellied Pacu (*Piaractus brachypomus*). *Mol Neurobiol* 61: 2620–2630. https://doi.org/10.1007/s12035-023-03700-5
56. Casaletto KB, Elahi FM, Bettcher BM, et al. (2017) Neurogranin, a synaptic protein, is associated with memory independent of Alzheimer biomarkers. *Neurology* 89: 1782–1788. https://doi.org/10.1212/WNL.0000000000004569
57. Vasung L, Abaci Turk E, Ferradal SL, et al. (2019) Exploring early human brain development with structural and physiological neuroimaging. *Neuroimage* 187: 226–254. https://doi.org/10.1016/j.neuroimage.2018.07.041

58. Subramanian L, Calcagnotto ME, Paredes MF (2020) Cortical malformations: Lessons in human brain development. *Front Cell Neurosci* 13: 576. https://doi.org/10.3389/fncel.2019.00576
59. Bick J, Nelson CA (2016) Early adverse experiences and the developing brain. *Neuropsychopharmacology* 41: 177–196. https://doi.org/10.1038/npp.2015.252
60. Lista S, Santos-Lozano A, Emanuele E, et al. (2024) Monitoring synaptic pathology in Alzheimer's disease through fluid and PET imaging biomarkers: a comprehensive review and future perspectives. *Mol Psychiatry* 29: 847–857. https://doi.org/10.1038/s41380-023-02376-6
61. Svirsky S, Henchir J, Li Y, et al. (2019) Neurogranin protein expression is reduced after controlled cortical impact in rats. *J Neurotrauma* 37: 939–949. https://doi.org/10.1089/neu.2019.6759
62. Garrido-García A, de Andrés R, Jiménez-Pompa A, et al. (2019) Neurogranin expression is regulated by synaptic activity and promotes synaptogenesis in cultured hippocampal neurons. *Mol Neurobiol* 56: 7321–7337. https://doi.org/10.1007/s12035-019-1593-3
63. Hwang H, Szucs MJ, Ding LJ, et al. (2021) Neurogranin, encoded by the schizophrenia risk gene NRGN, bidirectionally modulates synaptic plasticity via calmodulin-dependent regulation of the neuronal phosphoproteome. *Biol Psychiatry* 89: 256–269. https://doi.org/10.1016/j.biopsych.2020.07.014
64. Cheriyan VT, Alfaidi M, Jorgensen AN, et al. (2020) Neurogranin regulates eNOS function and endothelial activation. *Redox Biol* 34: 101487. https://doi.org/10.1016/j.redox.2020.101487
65. Lin H, Zhang J, Dai Y, et al. (2023) Neurogranin as an important regulator in swimming training to improve the spatial memory dysfunction of mice with chronic cerebral hypoperfusion. *J Sport Heal Sci* 12: 116–129. https://doi.org/10.1016/j.jshs.2022.01.008
66. Gribaudo S, Saraulli D, Nato G, et al. (2021) Neurogranin regulates adult-born olfactory granule cell spine density and odor-reward associative memory in mice. *Int J Mol Sci* 22: 4269. https://doi.org/10.3390/ijms22084269
67. Cardozo LP, de Lima BQI, Maciel MAE, et al. (2019) Synaptic elimination in neurological disorders. *Curr Neuropharmacol* 17: 1071–1095. https://doi.org/10.2174/1570159X17666190603170511
68. Ordyan M, Bartol T, Kennedy M, et al. (2020) Interactions between calmodulin and neurogranin govern the dynamics of CaMKII as a leaky integrator. *PLoS Comput Biol* 16: e1008015. https://doi.org/10.1371/journal.pcbi.1008015
69. Putkey JA, Hoffman L, Berka V, et al. (2024) Neurogranin modulates the rate of association between calmodulin and target peptides. *Biophys J* 123: 1676–1689. https://doi.org/10.1016/j.bpj.2024.05.010
70. O'Day DH (2020) Calmodulin binding proteins and Alzheimer's disease: Biomarkers, regulatory enzymes and receptors that are regulated by calmodulin. *Int J Mol Sci* 21: 7344. https://doi.org/10.3390/ijms21197344
71. Moradi F, Copeland EN, Baranowski RW, et al. (2020) Calmodulin-binding proteins in muscle: A minireview on nuclear receptor interacting protein, neurogranin, and growth-associated protein 43. *Int J Mol Sci* 21: 1016. https://doi.org/10.3390/ijms21031016
72. Dulewicz M, Kulczyńska-Przybik A, Słowik A, et al. (2021) Neurogranin and neuronal pentraxin receptor as synaptic dysfunction biomarkers in Alzheimer's disease. *J Clin Med* 10: 4575. https://doi.org/10.3390/jcm10194575

73. Poejo J, Salazar J, Mata AM, et al. (2021) The relevance of amyloid β-calmodulin complexation in neurons and brain degeneration in Alzheimer's disease. *Int J Mol Sci* 22: 4976. https://doi.org/10.3390/ijms22094976
74. Kumar V, Chichili VPR, Zhong L, et al. (2013) Structural basis for the interaction of unstructured neuron specific substrates neuromodulin and neurogranin with calmodulin. *Sci Rep* 3: 1392. https://doi.org/10.1038/srep01392
75. Szklarczyk D, Kirsch R, Koutrouli M, et al. (2023) The STRING database in 2023: Protein–protein association networks and functional enrichment analyses for any sequenced genome of interest. *Nucleic Acids Res* 51: D638–D646. https://doi.org/10.1093/nar/gkac1000
76. Nazir FH, Camporesi E, Brinkmalm G, et al. (2021) Molecular forms of neurogranin in cerebrospinal fluid. *J Neurochem* 157: 816–833. https://doi.org/10.1111/jnc.15252
77. Joglekar AP (2022) A cell-type centric view of alternative splicing in the mammalian brain. Available form: https://research.weill.cornell.edu/about-us/news-updates/cell-type-centric-view-alternative-splicing-mammalian-brain
78. Beghi S, Furmanik M, Jaminon A, et al. (2022) Calcium signalling in heart and vessels: Role of calmodulin and downstream calmodulin-dependent protein kinases. *Int J Mol Sci* 23: 16139. https://doi.org/10.3390/ijms232416139
79. O'Day DH (2022) Calmodulin binding domains in critical risk proteins involved in neurodegeneration. *Curr Issues Mol Biol* 44: 5802–5814. https://doi.org/10.3390/cimb44110394
80. Zhang H, Jiang X, Ma L, et al. (2022) Role of Aβ in Alzheimer's-related synaptic dysfunction. *Front Cell Dev Biol* 10: 964075. https://doi.org/10.3389/fcell.2022.964075
81. Abi-Dargham A, Moeller SJ, Ali F, et al. (2023) Candidate biomarkers in psychiatric disorders: State of the field. *World Psychiatry* 22: 236–262. https://doi.org/10.1002/wps.21078
82. Blennow K, Diaz-Lucena D, Zetterberg H, et al. (2019) CSF neurogranin as a neuronal damage marker in CJD: A comparative study with AD. *J Neurol Neurosurg Psychiatry* 90: 846–853. https://doi.org/10.1136/jnnp-2018-320155
83. Rabbito A, Dulewicz M, Kulczyńska-Przybik A, et al. (2020) Biochemical markers in Alzheimer's disease. *Int J Mol Sci* 21: 1989. https://doi.org/10.3390/ijms21061989
84. Mavroudis IA, Petridis F, Chatzikonstantinou S, et al. (2020) A meta-analysis on CSF neurogranin levels for the diagnosis of Alzheimer's disease and mild cognitive impairment. *Aging Clin Exp Res* 32: 1639–1646. https://doi.org/10.1007/s40520-019-01326-z
85. Siddappaji KK, Gopal S (2021) Molecular mechanisms in Alzheimer's disease and the impact of physical exercise with advancements in therapeutic approaches. *AIMS Neurosci* 8: 357–389. https://doi.org/10.3934/Neuroscience.2021020
86. Vaughn MN, Winston CN, Levin N, et al. (2022) Developing biomarkers of mild traumatic brain injury: Promise and progress of CNS-derived exosomes. *Front Neurol* 12: 698206. https://doi.org/10.3389/fneur.2021.698206
87. Kivisäkk P, Carlyle BC, Sweeney T, et al. (2022) Increased levels of the synaptic proteins PSD-95, SNAP-25, and neurogranin in the cerebrospinal fluid of patients with Alzheimer's disease. *Alzheimers Res Ther* 14: 58. https://doi.org/10.1186/s13195-022-01002-x
88. Kent SA, Spires-Jones TL, Durrant CS (2020) The physiological roles of tau and Aβ: Implications for Alzheimer's disease pathology and therapeutics. *Acta Neuropathol* 140: 417–447. https://doi.org/10.1007/s00401-020-02196-w

89. Bouwman FH, Frisoni GB, Johnson SC, et al. (2022) Clinical application of CSF biomarkers for Alzheimer's disease: From rationale to ratios. *Alzheimers Dement* 14: e12314. https://doi.org/10.1002/dad2.12314
90. Saunders TS, Gadd DA, Spires-Jones TL, et al. (2022) Associations between cerebrospinal fluid markers and cognition in ageing and dementia: A systematic review. *Eur J Neurosci* 56: 5650–5713. https://doi.org/10.1111/ejn.15656
91. Lepeta K, Lourenco MV, Schweitzer BC, et al. (2016) Synaptopathies: Synaptic dysfunction in neurological disorders–A review from students to students. *J Neurochem* 138: 785–805. https://doi.org/10.1111/jnc.13713

AIMS Press