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Protective Effects of Nicotinamide Adenine Dinucleotide and Related Precursors in Alzheimer's Disease: A Systematic Review of Preclinical Studies

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Abstract

Data from preclinical studies propose nicotinamide adenine dinucleotide (NAD⁺) as a neuroprotective and bioenergetics stimulant agent to treat Alzheimer's disease (AD); however, there seems to be inconsistency between behavioral and molecular outcomes. We performed this systematic review to provide a better understanding of the effects of NAD⁺ in rodent AD models and to summarize the literature.

Studies were identified by searching PubMed, EMBASE, Scopus, Google Scholar, and the reference lists of relevant review articles published through December 2020. The search strategy was restricted to articles about NAD⁺, its derivatives, and their association with cognitive function in AD rodent models. The initial search yielded 320 articles, of which 11 publications were included in our systematic review.

Based on the primary outcomes, it was revealed that NAD⁺ improves learning and memory. The secondary endpoints also showed neuroprotective effects of NAD⁺ on different AD models. The proposed neuroprotective mechanisms included, but were not limited to, the attenuation of the oxidative stress, inflammation, and apoptosis, while enhancing the mitochondrial function.

The current systematic review summarizes the preclinical studies on NAD⁺ precursors and provides evidence favoring the pro-cognitive effects of such components in rodent models of AD.

Keywords Alzheimer's disease · Cognition · Neuroprotection · Nicotinamide · Rodent

Introduction

In the current aging society, the number of patients suffering from Alzheimer's disease (AD) is rapidly increasing, and without therapeutic breakthroughs is estimated to reach 80 million by 2040 (Li et al. 2016). As an irreversible chronic neurodegenerative disease, AD is characterized by progressive cognitive deficits, memory loss, and personality changes taking place with advancing age (Dong et al. 2019). Neuropathological hallmarks of AD are mainly associated with the accumulation of extracellular senile amyloid-beta (A β) plaques and intracellular neurofibrillary

tangles (NFTs), consisting of hyperphosphorylated tau proteins (Sadigh-Eteghad et al. 2015). Studies have shown that extensive distribution of A β oligomers and NFTs within neurons is associated with the generation of oxygen free radicals and the inhibition of synaptic transmission and long-term potentiation (LTP) (Lesné et al. 2006; Wilkinson and Landreth 2006). Synaptic dysfunction, and especially the degeneration of cholinergic neurons, also leads to cognitive impairment and deficits in memory and learning and in emotional resilience (Chen and Mobley 2019). Oxidative stress, apoptosis, inflammation, autophagy, and mitochondrial dysfunction have also been widely shown to be involved in the progress of the disease (Bostancikloğlu 2019; Butterfield and Halliwell 2019). Thus, it seems that novel multi-targeted treatments are needed to prevent or slow disease progression.

Nicotinamide adenine dinucleotide (NAD⁺) is a crucial cofactor involved in a wide spectrum of biological functions including oxidative phosphorylation, mitochondrial function and bioenergetics, cell proliferation, and calcium homeostasis (Wei et al. 2017). Also, NAD⁺ is essential for appropriate

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neuronal function and survival in the central nervous system (Fang et al. 2014). NAD⁺ enters mammalian mitochondria via two transporters, SLC25A51 and SLC25A52 (Luongo et al. 2020). It has been shown that during the process of pathological aging and neurodegenerative disease, mitochondrial dysfunction is associated with a reduction in NAD⁺, which in turn prevents cellular respiration, leading to reduced adenosine triphosphate (ATP) levels and neuronal death (Hosseini et al. 2019). Since neurons have a high demand for energy and are very sensitive to NAD⁺ deficiency, maintaining intracellular levels of NAD⁺ through its precursors such as nicotinamide riboside (NR), nicotinamide mononucleotide (NMN), and nicotinamide (NAM) is essential. In addition, these agents could be therapeutic candidates for metabolic dysfunction in age-related neurodegenerative diseases (Liu et al. 2008). Several studies have examined the possible therapeutic effects of NAD⁺ precursors in preclinical models of AD (Gong et al. 2013; Long et al. 2015). In the current systematic review, we summarize the effects of NAD⁺ precursors in preclinical studies of AD and further discuss the neuroprotective and pro-cognitive effects of NAD⁺ precursors.

Methods

Search Strategy

The literature was systematically searched from four independent databases, i.e. PubMed, EMBASE, Scopus, and Google Scholar, for studies published no later than December 2020. Specific Medical Subject Headings (MeSH) and keywords that were searched included the following: [(Alzheimer Disease) OR (AD)] AND [(nicotinamide) OR (nicotinamide-beta-ribose) OR (niacin) OR (Nicotinamide adenine dinucleotide) OR (NAD⁺) OR (Nicotinamide Mononucleotide)] AND [(rodent) OR (rat) OR (mice) OR (mouse) OR (Rattus)].

The search was limited to original English-language articles, with no date restrictions. The reference lists of all included studies were further searched to avoid missing any relevant publications.

Inclusion and Exclusion Criteria

Original research articles published in English having an administration regimen of NAD⁺ precursors were included. We did include all dosages, administration routes, and administration time intervals of the drugs.

Non-rodent models, ex pathogenesis and development or in vitro (primary culture or cell line) experiments,

non-relevant treatments, duplicate articles, reviews, theses, and books were excluded from the study.

Data Extraction and Quality Assessment

Based on the inclusion and exclusion criteria, two investigators independently screened the titles/abstracts and the full texts, where necessary, and disagreements were resolved by a third party, i.e. senior researcher. The articles were listed based on (i) authors and publication year; (ii) animal characteristics including species, sex, weight, and age; (iii) duration of treatment, dosage, and administration route; and (iv) the model used in the study (Table 1).

To examine the internal validity of the selected studies (e.g., selection, performance, detection, and attrition bias) and other study quality measures (e.g., reporting quality and power), a modified version of the Collaborative Approach to Meta Analysis and Review of Animal Data from Experimental Studies (CAMARADES) quality checklist was used. The items in the list comprised (i) publication in a peer-reviewed journal, (ii) randomization to treatment or control, (iii) allocation concealment, (iv) blinded assessment of an outcome, (v) statement of inclusion and exclusion of animals in the study, (vi) sample size calculation, (vii) statement of compliance with regulatory requirements, and (viii) statement of possible conflicts of interest.

Results

Study Selection

We evaluated the results of 11 studies investigating the effects of NAD⁺ precursors in the experimental AD models. A PRISMA [Preferred Reporting Items for Systematic Reviews and Meta-Analyses] flowchart and the study selection process are presented in Fig. 1. Overall, 247 articles were found after excluding the replicated studies. Of these, 205 articles were excluded based on the title and the abstract. Full texts of 42 articles were further read in order to assess their eligibility, excluding the non-relevant papers based on exclusion criteria. Finally, 11 qualified articles were included in the systematic review.

Quality of Included Studies

All investigated articles are published in peer-reviewed journals. A disclosure statement for conflict of interest was present in 91.67% of the studies. Thirty-three percent of the studies reported random allocation. Compliance with animal welfare regulations was reported in 83.34% of the

Table 1. Characteristics of the studies included in the systematic review

Authors & Year	AD model	Species	Sex Age (months)	Drug	Highest dose (mg/kg)	Duration of treatment (Days)	Route	Main findings
Liu et al. (2013)	3xTg-AD	Mice	NM 4	NAM	40	240	Drinking water	↑Cognitive function, NAD ⁺ levels and activation of Akt, ERKs, and CREB. Preserved mitochondrial integrity Improved autophagy-lysosome processing and brain bioenergetics ↓Aβ and p-Tau
Gong et al. (2013)	Tg2576-AD	Mice	NM 8	NR	250	90	Drinking water	↑PGC-1α and NAD ⁺ . ↓Cognitive deterioration and Aβ levels
Green et al. (2008)	3xTg-AD	Mice	NM 4	NA	200	120	Drinking water	↑Cognitive function and acetylated α-tubulin ↓p-Tau
Kim and Yang (2017)	Aβ ₁₋₄₂	Mice	Male 2	NA, NAM	NA, 20 NAM, 400	7	Oral	↓APP, NF-κB, and PS1 ↑Sirt1 expression
Bayrakdar et al. (2014)	Aβ ₁₋₄₂	Rat	Male NM	NA	500	7	i.p.	↑Bcl-2 ↓PARP-1, NF-κB, p53, and Bax levels, oxidative stress factors
Hou et al. (2018)	3xTg-AD	Mice	NM 16-18	NR	12 mM	90	Drinking water	↑Cognitive function, restored hippocampal synaptic plasticity, NAD ⁺ /NADH ratio, and SIRT3 activity ↓p-Tau, DNA damage, neuro-inflammation, and apoptosis
Long et al. (2015)	APP(swe)/PS1(ΔE9)	Mice	Male and female 2	NMN	100	14	s.c.	↓APP levels and mitochondrial fragmentation
Vakilinezhad et al. (2018)	STZ	Rat	Male NM	NA NA-PS-SLN _s	-	-	Oral i.p. i.v.	↑Memory and ↓p-Tau levels
Wang et al. (2016)	Aβ ₁₋₄₂	Rat	Male NM	NMN	500	10	i.p.	↑NAD ⁺ level, and energy metabolism ↓ROS accumulation, and inhibition of LTP

Table 1. (continued)

Authors & Year	AD model	Species	Sex Age (months)	Drug	Highest dose (mg/kg)	Duration of treatment (Days)	Route	Main findings
Xie et al. (2019)	APP/PS1	Mice	Male 3	NR	400	90	Oral	↑ Contextual fear memory ↓ Chronic neuroinflam- mation, A β accumulation and migration of astrocytes
Yao et al. (2017)	APP/PS1	Mice	NM 6	NMN	100	14	s.c.	↓ Cognitive impairments, A β , JNK activation, syn- aptic loss, and inflammatory responses ↑ SYP and PSD- 95 levels

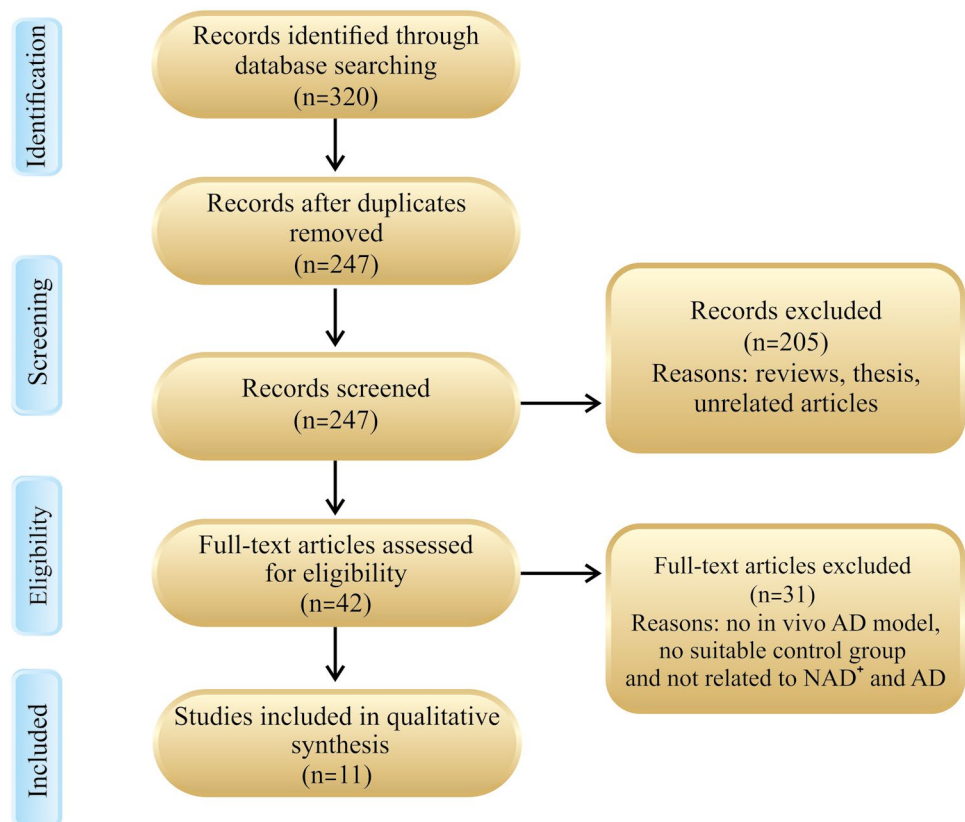
MWM Moris water maze, *NOR* novel object recognition, *OF* open field, *EMP* elevated plus maze, *FST* forced swimming test, *FC* fear conditioning, *PA* passive inhibitory avoidance, *NM* not mentioned, *NR* nicotinamide riboside, *NMN* nicotinamide mononucleotide, *NAM* nicotinamide, *NAD⁺* nicotinamide adenine dinucleotide

studies. The sample size calculation was mentioned in none of the publications. The model and animal exclusion criteria and blind outcome assessment were reported in 16.67% and 8.33% of the papers, respectively (Fig. 2).

AD Models in Included Studies

The 11 included studies involved five main types of AD models: (i) amyloid precursor protein (APP), (ii) presenilin 1

Fig. 1 PRISMA flowchart and the study selection process of the systematic review



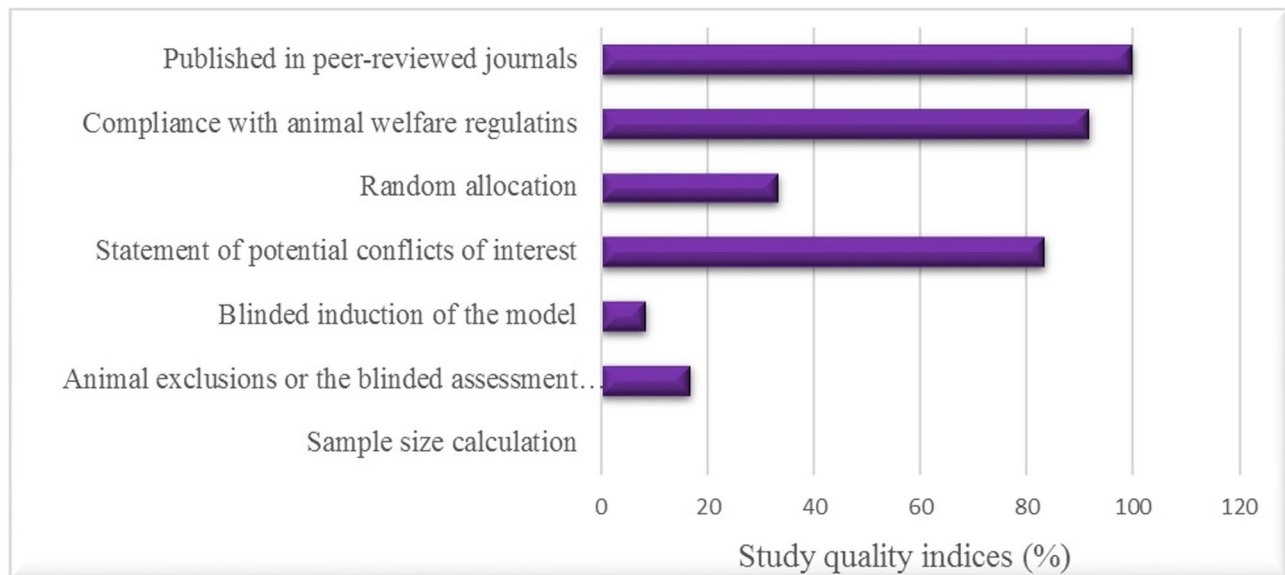


Fig. 2 Evaluation of the included studies based on the modified CAMARADES quality checklist

(PS1), (iii) tau transgenic models, (iv) streptozotocin (STZ), and (v) $A\beta_{1-42}$ -induced surgical models.

Primary Outcomes

Behavioral Tests

Among the included studies, eight investigated the effects of NAD^+ precursors on behavioral outcomes (Table 2). Tasks for behavioral studies included the Morris water maze (MWM), elevated plus maze (EPM), Y-maze, novel object recognition (NOR), passive avoidance (PA), fear conditioning (FC), and open field (OF) tests. These tasks assess learning, memory, and depression- and anxiety-like behaviors in various experimental models of AD.

The results of a study by Liu et al. (2013) showed that NAM administration for 8 months significantly reduced goal latencies and path lengths, and markedly increased the time spent in the target quadrant in the MWM task. In the OF test, mice treated with NAM exhibited more vertical counts and stereotyped movements. However, no significant relation was established for the distance traveled and the ambulatory counts between NAM-treated 3xTg-AD and control groups (Liu et al. 2013). In another study, NR supplementation was revealed to improve recognition memory performance in Tg2576 mice, as assessed by the NOR test (Gong et al. 2013). According to Green et al. (2008), NAM treatment via an oral route in AD mice led to a noticeable improvement in spatial memory and prevented contextual fear memory deficits. Also, in the same study, no difference was found between NA-treated 3xTg-AD animals and the vehicle group

Table 2 The effects of NAD^+ precursors on behavioral outcomes in different tasks in the studies

	MWM	NOR	OF	EPM	FC	Y-maze	PA
Liu et al. (2013)	+		+				
Gong et al. (2013)		+					
Green et al. (2008)	+	NS					+
Hou et al. (2018)	+	+	NS	+	+	+	
Vakilinezhad et al. (2018)	+						
Wang et al. (2016)	+		NS				
Yao et al. (2017)	+						+
Xie et al. (2019)		NS	NS	NS	+	NS	

"+" significant positive effects, NS nonsignificant effects, MWM Morris water maze, NOR novel object recognition, OF open field, EPM elevated plus maze, FST forced swimming test, FC fear conditioning, PA passive inhibitory avoidance

in the NOR test. As reported by Hou et al., AD induction impairs spatial learning, recognition, and working memory in animals, while NR treatment reverses such deficits (Hou et al. 2018). NR was also found to increase cued and context freezing times and decrease anxiety-like behavior in AD mice. In a study by Vakilinezhad et al. (2018), NAM-loaded phosphatidylserine-solid lipid nanoparticles (PS-SLN) alleviated spatial learning and memory deficits in AD rats. In other studies, NMN treatment counteracted learning deficits and ameliorated MWM performance (Yao et al. 2017; Zhang et al. 2016). According to Xie et al., supplementation of NR improved contextual fear memory, but not cue fear memory, in APP/PSA1 mice (Xie et al. 2019). Also, there was no significant difference in anxiety behavior and short-term memory between the NR and the control group in APP/PSA1 mice.

Secondary Outcomes

Neuroprotective Mechanisms

A β - and Tau-Related Mechanisms

Among the 11 studies included in the review, eight investigated the effects of NAD⁺ precursors on A β plaque aggregation and/or tau hyperphosphorylation. Liu et al. (2013) illustrated that NAM treatment decreased the levels of p-tau and A β accumulation in the CA1 region of the hippocampus (HIP), subiculum, and the cerebral cortex in 3xTg mice. It was also reported that administration of NR for 3 months reduced A β production (Gong et al. 2013). Although Green and colleagues did not observe any effect of NAM treatment on A β pathology, they reported decreased Thr231-p-tau levels in 3xTg-AD mice (Green et al. 2008).

Similarly, evidence from the study by Hou et al. indicated that NR-treated AD mice demonstrated a decreased level of p-tau pathology, but no impact was observed on A β accumulation (Hou et al. 2018). Another study revealed that NAM prevented tau hyperphosphorylation in a STZ-induced rat model of AD (Vakilinezhad et al. 2018). Xie et al. demonstrated that an NR regimen inhibited A β accumulation while also blocking astrocyte migration towards zones of A β accumulation (Xie et al. 2019). A marked reduction in the levels of A β ₁₋₄₀, A β ₁₋₄₂ and APP expression was observed in the cortex and HIP of NMN-treated transgenic AD mice (Yao et al. 2017). Kim and Yang reported that NAM pretreatment decreased the expression of APP and PS1 in brain tissue (Kim and Yang 2017).

Under physiological conditions, microtubule-associated protein 2 (MAP2) is located predominantly in the cell body and dendrites, while tau is found mostly in axons. MAP2 is

considered a main component of the neuronal cytoskeleton, with a key role in different stages of nervous system development, formation, and regeneration. Tau triggers tubulin polymerization and induces microtubule stabilization (Manczak et al. 2018). Tau is phosphorylated by two major kinases, glycogen synthase kinase 3 (GSK3) and cyclin-dependent kinase 5 (cdk5) (Hu et al. 2011). The accumulation of phosphorylated tau in the hippocampal area is responsible for the loss of dendritic protein MAP2, which in turn results in neuronal death, and consequently impaired learning and memory (Kandimalla et al. 2018). Based on the results of the study by Green et al., oral administration of NA cannot modulate the activity of GSK3 β and cdk5, but can enhance learning and memory via the upregulation of structural and synaptic proteins (Green et al. 2008). In fact, decreased acetylated α -tubulin was observed in tangle-bearing neurons in the AD brain. Interestingly, NA intervention was shown to increase acetyl- α -tubulin levels significantly; however, it had no effect on the steady-state levels of total monomeric or dimeric α -tubulin. This study also found a significant increase in MAP2c in NA-treated AD mice compared to the vehicle group (Green et al. 2008).

Although MAP2 shares highly homologous carboxyl-terminal sequences with tau, these microtubules have different fates in the course of NFT formation in the AD brain. It has been demonstrated that tau forms filaments when aggregated, while MAP2 aggregates to granules. One study found that MAP2 was not implicated in the growth process of NFT in the AD brain (Xie et al. 2015).

Anti-inflammatory Effects

Among the included studies, five articles assessed the anti-inflammatory effect of NAD⁺ precursors in AD mice. Inflammation in neurodegenerative diseases is characterized by activated microglia, reactive astrocytes, and increased release of inflammatory cytokines (Kim et al. 2004; Waetzig et al. 2005). Glial fibrillary acidic protein (GFAP) and ionized calcium-binding adaptor molecule 1 (IBA1) are specific markers for activated astrocytes and microglia, respectively (Kaur et al. 2015). Hou et al. provided evidence that NR reduced GFAP and IBA1 in both the cortex and HIP regions and normalized the levels of pro-inflammatory and inflammatory cytokines including interleukin (IL)-1 α , IL-1 β , tumor necrosis factor (TNF)- α , MCP-1, MIP-1 α , and RANTES in AD mice (Hou et al. 2018). Another study (Xie et al. 2019) showed that supplementation of NR decreased the density of GFAP-positive cells in the HIP of APP/PS1 mice, but had no effect on the density of IBA1-positive cells in the cortex and hippocampal dentate gyrus. Also, Yao et al. found that treatment with NMN inhibited c-Jun N-terminal kinase (JNK) activation, leading to suppression of glial cell activation. This further

reduced the levels of proinflammatory cytokines such as IL-6, IL-1 β , and TNF- α (Yao et al. 2017). NAM treatment was also shown to inhibit poly (ADP-ribose) polymerase-1 (PARP-1) overactivation (Turunc Bayrakdar et al. 2014) and reduce the mRNA and protein levels of nuclear factor kappa B (NF- κ B) expression (Kim and Yang 2017; Turunc Bayrakdar et al. 2014).

Mitochondrial Function and Bioenergetics

Mitochondrial dysfunction is a prominent feature of the AD brain, and leads to morphological and functional abnormalities and limits electron transport chain and ATP production (Bonda et al. 2010). A decreased level of ATP reduces the neuron's ability to maintain ionic gradients, which further hinders the production and propagation of action potentials, and thus, neurotransmission (Butterfield and Halliwell 2019). Mitochondrial morphology is modulated by a dynamic balance between fission and fusion. Mitochondrial fusion is regulated by protein optic atrophy 1 (Opa1) and mitofusin proteins (Mfn1 and Mfn2), and its fission is regulated by dynamin-related protein (Drp1) and fission protein 1 (FIS1) (Zhu et al. 2013). It was demonstrated that the balance between mitochondrial fission and fusion is impaired in AD (Wang et al. 2009). Long et al. found that the rate of mitochondrial fission was higher in transgenic AD models than that in control animals. Mitochondrial dynamics were shown to be improved in NMN-treated AD mice (Long et al. 2015).

Four studies also investigated the effects of NAD⁺ precursor supplementation on the levels of NAD⁺, and all reported increased levels of NAD⁺ in AD models (Gong et al. 2013; Hou et al. 2018; Liu et al. 2013; Zhang et al. 2016).

The study by Liu et al. (2013) revealed that NAM treatment normalized mitochondrial dynamics and improved autophagy-lysosome procession via enhanced lysosome/autolysosome acidification to reduce autophagosome accumulation. NAM treatment was also found to activate neuroplasticity-related kinases (p-Akt, MAPK/ERK42, and cyclic AMP response element-binding protein [CREB]) and elevate sirtuin 1 (SIRT1) protein levels in the brains of AD mice. Gong et al. (2013) reported that NR improved synaptic plasticity and facilitated proliferator-activated receptor- γ coactivator 1 α (PGC-1 α) and β -secretase 1 degradation. Furthermore, it promoted the expression of energy metabolism genes citrate synthase, cytochrome c subunit Vic, aconitase, pyruvate dehydrogenase kinase 3, phosphoglycerate kinase, and glucose phosphate isomerase 1. Kim and Yang showed that the administration of NAM in AD mice increased the expression of SIRT1 (Kim and Yang 2017).

Oxidative Stress and Synaptic Factors

Oxidative stress is an imbalance between the production of reactive oxygen species (ROS)/reactive nitrogen species (RNS) and the counteracting antioxidant mechanisms. It is a major contributor to the pathogenesis and development of AD (Ton et al. 2020). Turunc Bayrakdar et al. (2014) reported that NA markedly reduced oxidative stress elements including protein carbonyls, ROS, and malondialdehyde (MDA), and increased antioxidant enzyme activity in the rat brain. This study also found that mRNA and protein levels of apoptosis markers were decreased in the HIP and cortex of AD rats treated with NA (Turunc Bayrakdar et al. 2014).

NAD⁺ precursor administration may affect the synaptic factors in the brain of AD animals. It was shown that the expression of MAP2c was increased after NA administration in AD animals (Green et al. 2008). Moreover, Yao et al. reported that NMN ameliorated synaptic loss and increased the levels of postsynaptic density protein-95 (PSD-95) and synaptophysin (SYP) expression in the cortex and HIP of AD mice (Yao et al. 2017). In another study, Wang et al. (Zhang et al. 2016) revealed that treatment of AD rats with NMN increased LTP, restored the level of ATP, and decreased ROS and superoxide levels in the mitochondria (Fig. 3).

Discussion

This systematic review supports the efficacy of NAD⁺ precursors for improving both behavioral and neuroprotective outcomes in animal models of AD. NAD⁺ is an energy substrate and cofactor for several enzymes, including sirtuins, ADP-ribosyl cyclase (CD38), PARP-1, and NAD⁺-dependent dehydrogenases (Davila et al. 2018; Ying 2008). NAM, NAD⁺, and nicotinic acid are incorporated into cells using a transporter. NMN is converted to NR by CD73 5'-ectonucleotidase in the extracellular space. Cell entrance of NR can be performed through nucleoside transporters (Mericskay 2016). Recently, a new NMN-specific transporter, i.e. Slc12a8, was identified in mammals (Grozio et al. 2019).

Because intracellular NAD⁺ is an essential mediator of cell survival, (Hosseini et al. 2019), its restoration can play an important role in reducing degeneration. A growing body of evidence has shown that NAD⁺ plays an important role in brain metabolism, and has a crucial influence on neurotransmission, synaptic plasticity, learning, and memory (Wang et al. 2015). As demonstrated by the present review, treatment with NAD⁺ precursors restored NAD⁺ levels in the brain of rodent models of AD (Gong et al. 2013; Hou et al. 2018; Liu et al. 2013; Zhang et al. 2016). Also, the administration of precursors increased rodents' spatial memory (Green et al. 2008; Hou et al. 2018; Liu et al. 2013; Vakilinezhad

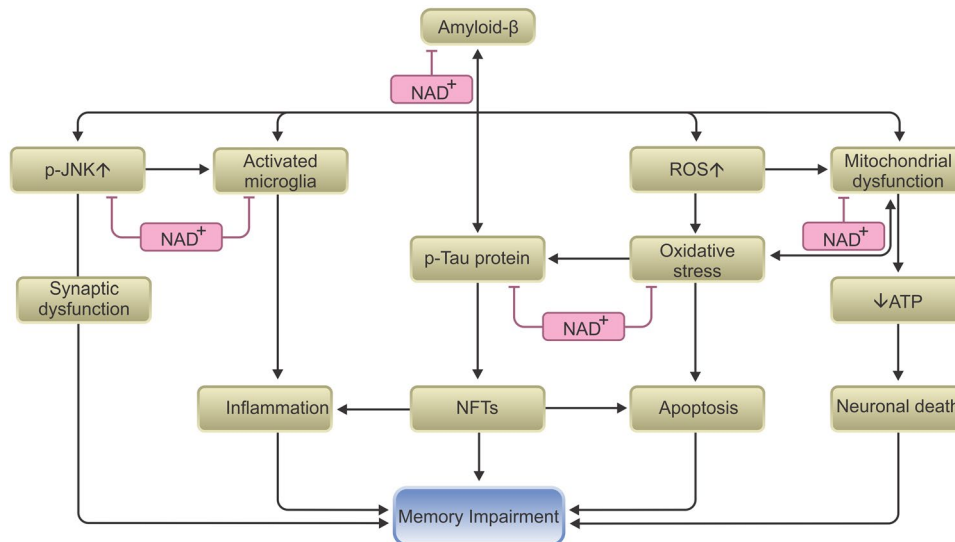


Fig. 3 Schematic diagram showing the molecular mechanisms involved in the pathogenesis of Alzheimer's disease (AD) and the role played by NAD^+ as a neuroprotective regimen. Amyloid-beta ($\text{A}\beta$) triggers a cascade of events including mitochondrial dysfunction, oxidative stress, p-tau, and activation of microglia and JNK. Oxidative stress disrupts neuronal mitochondrial function, increases p-tau and $\text{A}\beta$ production, apoptosis, and neuroinflammation. Hyperphospho-

rylated tau protein is the main constituent of the NFTs. These events lead to neural dysfunction, followed by neural death, and finally, memory impairment. NAD^+ exerts neuroprotective effects through the modulation of multiple points in the complex AD cascade. NAD^+ : nicotinamide adenine dinucleotide, NFTs: neurofibrillary tangles

et al. 2018; Yao et al. 2017; Zhang et al. 2016), contextual learning, and freezing behavior (Hou et al. 2018; Xie et al. 2019). These treatments were also shown to alleviate AD-related dementia by inhibiting $\text{A}\beta$ production and reducing tau phosphorylation (Gong et al. 2013; Liu et al. 2013; Xie et al. 2019).

As mentioned above, neuroinflammation is known as a major contributor to AD pathogenesis (Kim et al. 2004; Waetzig et al. 2005). $\text{A}\beta$ fragments are able to induce activation of JNK, leading to phosphorylation of both tau and APP (Minogue et al. 2003; Vogel et al. 2009). Clinical studies have reported increased expression of phosphorylated JNK in the brains of AD patients (Killick et al. 2014; Zhu et al. 2001). It has been demonstrated that JNK promotes neuroinflammatory processes in AD by activating microglial cells (Mehan et al. 2011). The activated form of JNK is involved in beta-site APP-cleaving enzyme (BACE1) expression (Guglielmo et al. 2011; Rahman et al. 2012), which plays a role in the generation of $\text{A}\beta$. Also, the abnormal activation of microglia leads to the generation of proinflammatory mediators, neuronal apoptosis, inhibition of neurogenesis, and decreased hippocampal volume, all of which are associated with AD (Wang and Colonna 2019). A recent study showed that the downregulation of JNK3 genetically led to a decrease in $\text{A}\beta$ levels and ameliorated cognitive impairment (Yoon et al. 2012). In addition, the hyperactivation of PARP-1, a nuclear enzyme, and

the accumulation of poly (ADP-ribose) (PAR) have been proven to exist in the brains of AD patients (Martire et al. 2015). The pathological activation of PARP-1 promotes cell death via mechanisms involving NAD^+ depletion. Therefore, PARP-1 knockdown or its pharmacological inhibition can notably increase cell survival partially by NAD^+ modulation (Mandir et al. 2000). Collectively, the results of the included studies show that NAD^+ mediators can prevent $\text{A}\beta$ -induced neuroinflammation by inhibiting JNK, PARP-1, and glial cell activation (Hou et al. 2018; Rahman et al. 2012; Turunc Bayrakdar et al. 2014; Yao et al. 2017), suggesting anti-inflammatory effects of NAD^+ . PSD-95 and SYP proteins play critical roles in synapse maturation and synaptic plasticity (Popugaeva et al. 2017). JNK activation triggers $\text{A}\beta$ -induced synaptic loss (Costello and Herron 2004; Sclip et al. 2014); in contrast, administration of precursors such as NMN elevates the expression of synaptic proteins via inhibition of JNK (Yao et al. 2017).

Increasing evidence suggests that oxidative stress triggers the generation of $\text{A}\beta$, p-tau, and mitochondrial dysfunction. Ultimately, the pathological cascade of events results in neuronal damage and memory impairment (Cassidy et al. 2020). Alterations in membrane phospholipid composition increase the levels of MDA, which is a known lipid peroxidation marker. This has been frequently reported in the brains of experimental AD models and cerebrospinal fluid of AD patients (Sultana and Butterfield 2010; Sultana

et al. 2006). The administration of NAD⁺ has been shown to decrease cell death induced by oxidative stress (Alano et al. 2004). Also, activation of NAD⁺ has been associated with a reduction in amyloid toxicity in the brains of the AD animal models (Kim et al. 2007). Furthermore, studies on AD animals have reported a reduction in ROS levels following NA or NMN treatment (Turunc Bayrakdar et al. 2014; Zhang et al. 2016). In the study by Turunc Bayrakdar et al., NA increased antioxidant enzyme activity and decreased MDA in the prefrontal cortex and HIP (Turunc Bayrakdar et al. 2014).

Aβ oligomers disrupt the brain bioenergetics system via interference with mitochondrial function (Onyango et al. 2016). Mitochondrial dynamics (fission and fusion) are essential for the maintenance of mitochondrial integrity, energy metabolism, inhibition of ROS production, and cellular apoptosis (Wang et al. 2017). Mitochondrial fission disassociates functional forms from damaged mitochondria via mitophagy (Wang et al. 2009). In AD, Aβ overproduction has been linked to extreme mitochondrial fragmentation (fission), which leads to mitochondrial dysfunction as evidenced by the loss of ATP synthesis and increased ROS that eventually results in cell death (Flannery and Trushina 2019; Zhu et al. 2013). Long and colleagues showed that mitochondrial dynamics were impaired in neurons affected by AD, while NMN therapy improved the balance of fission and fusion in the affected neurons (Long et al. 2015). Several studies demonstrated that PGC-1α, the primary regulator of energy metabolism, facilitates the degradation of the BACE1 protein (Gong et al. 2010; Kwak et al. 2011). It was also shown that NR treatment upregulates the expression of PGC-1α and promotes BACE1 degradation in the AD brain (Gong et al. 2013). The evidence further suggests that the silencing of PGC-1α can eradicate the effects of NR on BACE1 degradation (Gong et al. 2013). CREB and SIRT1 exhibit regulatory effects on the genes involved in neuronal survival and function in an Aβ-induced oxidative stress condition (Pugazhenthil et al. 2011; Su et al. 2014). Treatment with NAM also increases CREB and SIRT1 expression and activates Akt kinase, which is another possible mechanism for its pro-cognitive effects (Liu et al. 2013).

Conclusion

The data from the included studies showed that NAD⁺ precursors appeared to be an effective anti-AD strategy for preventing neuropathological and behavioral symptoms induced by AD in preclinical trials. Such favorable effects are possibly modulated by reducing central Aβ and tau levels and improving brain bioenergetics, inflammation, and oxidative stress regulation.

Authors' Contributions LH, JM, and SS-E designed the project. FP and HS-P performed the literature review and extracted data. LH and S S-E wrote the manuscript. S S-E supervised the work. JM and S S-E edited the final version.

Declarations

Consent for Publication The authors give the publisher permission to publish the work.

Competing Interests The authors declare they have no conflict of interest.

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