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EFFICACY OF NICOTINAMIDE MONONUCLEOTIDE SUPPLEMENTATION (NMN) IN BLOOD NICOTINAMIDE ADENINE DINUCLEOTIDE (NAD) FOR ANTI-AGING IN ADULTS: A SYSTEMATIC REVIEW

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ABSTRACT

Aging is a risk factor for various chronic diseases such as diabetes, dyslipidemia, metabolic, neurological, and cardiovascular disease, and other age-related disorders. Nicotinamide mononucleotide (NMN) is one of the endogenous intermediate molecules in nicotinamide adenine dinucleotide (NAD^+) biosynthesis, and its biological function is known to be related to an increase in NAD^+ concentration. NAD^+ levels decline and are critical for human health and longevity. The material and methods using a systematic review of RCT, with participants of healthy adults aged between 20-65 years old, agreed to take NMN supplementation compared to Placebo. The inclusion criteria are RCT studies regarding NMN administration with an outcome to prevent aging using primary objective measurement of blood nicotinamide adenine dinucleotide (NAD) concentration. Trial quality was assessed using the JBI checklist. We included six studies with an average duration of intervention between 10-12 weeks with a dosage of NMN 250-900mg/day. All six studies showed that NMN supplementation can significantly improve blood NAD concentration and cause only mild adverse effects during the trial. In conclusion, NMN supplementation has a significant clinical impact in preventing adult aging.

Keywords: *Adults, anti-aging, nicotinamide adenine dinucleotide, nicotinamide mononucleotide*

INTRODUCTION

The dramatic change in the global population from the decrease in the natality and the increase in the average life span led to the rapid growth of the proportion of citizens aged 60 and above. In 2022, the global population over age 65 was about 771 million (10%), and it is expected to increase (16%) to over 1.6 billion population in 2050.[1,2] The restraint of physiological deterioration in aging is an exceptional strategy to prevent metabolic and age-related disorders. The process of aging poses a risk factor for a variety of diseases, such as dyslipidemia, neurological and cardiovascular disease, and diabetes.[3] Some researchers still discover endogenous compounds that might attain longevity and productive and energetic lives.

Nicotinamide mononucleotide (NMN) is a naturally occurring molecule present in all tissues of mammals. However, it is a natural product that exists in most plants only in a small amount (~1mg/100gr food), such as broccoli, edamame, and cucumber.[4,5] NMN is a biologically active nucleotide naturally produced through the enzymatic conversion of a phosphate group and nicotinamide riboside (NR) by nicotinamide phosphoribosyl transferase (NAMPT). It additionally serves as an intermediary compound in the biosynthesis of nicotinamide adenine dinucleotide (NAD+), functioning as a crucial substrate for diverse enzymes that facilitate its enzymatic conversion to NAD+ in humans. It is a biological function related to the capability to elevate NAD+, a molecule vital for the existence of living organisms, to maintain the well-being of cells, including enhancing energy metabolism, anti-inflammatory, DNA repair, and anti-aging effects.[6] The decline in NAD levels is linked to characteristics of aging and diseases related to age, which are also critical for human health and longevity.[7]

Recent research primarily examines the impact of NMN solely within cellular or animal groups. Limited clinical trials in humans show that NMN supplementation can increase blood NAD concentration. Recent clinical trials have shown that NMN administration is secure and can address NAD deficiency. Hence, utilizing a systematic review study design, this study focused on assessing the effectiveness of the intake of NMN in enhancing NAD levels in the bloodstream for anti-aging purposes in adults. These data are essential to determine whether there are differences in the benefits and adverse effects of various dosages and duration of NMN supplementation to increase blood NAD concentration to maintain human longevity and health.

METHODS

Search strategy

This review performed an internet-based search for literature using journal databases in Google Scholar, Pubmed, and Science Direct. We used MeSH entry terms with keywords “nicotinamide adenine dinucleotide,” “NAD,” “nicotinamide mononucleotide,” “NMN,” “anti-aging,” “aging,” “human,” and “adults” using Boolean operator AND and OR to define the further issue.

Study eligibility

We identified studies meeting specific eligibility criteria through a PRISMA diagram, illustrated in Figure 1. During the first screening phase, we initially applied search strategy keywords to filter articles from an online database. Non-relevant or duplicated studies were subsequently excluded. Following this, we assessed the abstracts and examined the full-text versions to determine eligibility based on specific criteria. The criteria for inclusion are as follows: (1) Population: Studies involving middle-aged and older adult participants; (2) Intervention: Taking oral NMN supplementation; (3) Outcome: Evaluating the blood NAD concentration to prevent aging; (4) Study design: Only randomized control trials (RCT) to maintain the quality of this study. Instead, the exclusion criteria are research not conducted in humans (such as mice, rats, cell level, et cetera) and not using blood NAD measurement. These studies are review articles that do not use placebo-controlled study design, preclinical research, and articles not available in full text.

Study selection

Four reviewers collected relevant studies for inclusion, considering the topic and study design that aligned with our eligibility criteria. Initially, we examined the articles’ abstracts and then read the full-text versions. The chosen research underwent assessment for its evidence before being incorporated into the ultimate review. Each study was meticulously examined to distil the essential concepts from the literature (refer to Figure 1).

Study quality assessment

The study’s quality was evaluated using the Joanna Briggs Institute checklist for critical appraisal. Tailored checklists corresponding to each study design were engaged, with each checklist item carrying a single point, considered good if its score equals or surpasses half of the maximum total points [13].

Data synthesis

A comprehensive examination of studies on NMN supplementation for anti-aging in adults reveals a cohesive narrative analysis. This systematic review, being a qualitative report, sought to collect data regarding the impact of NMN supplementation on anti-aging in adults. We systematically conducted the narrative synthesis to determine the feasibility, effectiveness, and potential adverse effects of NMN supplementation for anti-aging in adults.

Result

At first, we identified 47 articles concerning NMN supplementation for anti-aging in adults. However, 13 did not meet our inclusion criteria for the study design because it did not use placebo-controlled consisted of review articles and preclinical research. The remaining 22 studies used animal subjects, five articles were inaccessible, and the final study did not employ blood NAD as a measurement. Ultimately, we found six articles and attempted to assess how NMN supplementation affects the aging process in adults.

We included six randomized, double-blind, placebo-controlled, multicentred, parallel-design clinical trials. The studies mainly came from Japan and India. There were 275 in the total sample, of which 150 participants (54%) were female subjects and 125 participants (36%) were male subjects involved in the studies. Most of the studies used male and female participants aged between 40 and 65, with average body mass index (BMI) and no underlying disease. The study by Igarashi *et al.* only used male participants, Okabe *et al.* only specifically used Japanese participants for their study, and the study by Yoshino *et al.* only used postmenopausal women with prediabetes, obese or overweight— the comprehensive features of the study are outlined in Table 1.

Based on each study's design, the Joanna Briggs Institute checklist evaluates the study's quality. All seven studies were randomized control trials. The interpretation of the checklist is that each item presents one point and counts as having good quality if the score is half or the maximum of the total point. If a study scores less than fifty percent of the maximum achievable points, it is categorized as low-quality. For Randomized Controlled Trials (RCT), the scoring range is 0-13, and the quality assessment involved five reviewers to ensure impartiality. All six of our studies were actively engaged and deemed good quality, with a total point range of 11-13.

In these six studies of our systematic review, the researchers gave several dosages of NMN supplementation. Four studies use an NMN dosage of 250mg/day, like a report conducted by Katayoshi *et al*., Igarashi *et al*., Okabe *et al*., and Yoshino *et al*.,[8–11] The other two studies' dosages are varied from 300mg to 900 mg per day.12,13 Interventions from the comparator or control groups all use a placebo. The intervention time varied between 10 weeks, 12 weeks, and 60 days.[8–13]

All the sixth studies evaluated the blood NAD concentration as the primary objective. Yin Li et al.'s study involved collecting blood samples on days 0, 30, and 60 during participants' visits to the trial centers.[13] The other study by Huang H measured the blood NAD concentration at days 30 and 60,[12] study by Igarashi *et al.* measured the blood NAD levels at the 6–12-week visit,[11] Okabe *et al.* evaluated the blood NAD level every four weeks,[9] Yoshino *et al.* and Katayoshi *et al.* only measured the blood NAD concentration once at weeks 10th and 12th, respectively.[8,10] In all six studies, it was evident that NMN supplementation led to a significant rise in blood NAD concentration.[8–13]

DISCUSSION

Effectively handling contagious illnesses throughout the 20th century substantially raised the average life expectancy in numerous nations. Populations over 65 years old worldwide is projected to be more than twice that of children under five. With the expanding aging demographic, there is an increasing prevalence of age-related conditions like hypertension, diabetes mellitus, atherosclerosis, osteoarthritis, Alzheimer's disease, and cancer.[14] About twelve hallmarks contribute to the aging process and determine the phenotype of aging and longevity.[15,16] To manage all these features, the medical practice of age management has greatly expanded worldwide. Recommendations for mitigating aging and prolonging a healthy lifespan may include medications, nutritional supplements, hormone therapies, exercise programs, and other treatments. Among various anti-aging healthcare products, NMN has drawn considerable interest in Europe, North America, and China over the past decade. NMN is an outstanding primary precursor of nicotinamide adenine dinucleotide (NAD+), an amide form of 5'- phosphoribosyl-pyrophosphate (PRPP) by nicotinamide phosphoribosyl transferase (NAMPT) and vitamin B3, a crucial coenzyme for fundamental cellular physiological functions such as aging, metabolism, DNA repair, cell death, gene expression, showing anti-aging effects.[17]

From our analysis in this systematic review, all six studies demonstrated a notable elevation in NAD levels in the bloodstream concentration using daily NMN supplementation.[8–13] A study by Yin *et al.* showed that the oral intake of NMN at a dosage of 900 mg/day for 60 days was deemed safe and well-tolerated.[13] Otherwise, a study by Katayoshi *et al.* stated that a 12-week of 250mg NMN supplementation was beneficial and safe to stimulate NAD metabolism in seemingly healthy individuals in their middle years.[8] Huang H proved an increase in NAD^+/NADH levels by 11.3% in serum at day 30 and 38% by the end of day sixtieth. There were no noteworthy irregularities in safety laboratory tests

that held clinical significance. Both groups were observed, suggesting the Uthever (NMN) supplement is safe.[12] Both studies by Igarashi *et al.* and Yoshino *et al.* showed that at weeks tenth and twelfth, regular oral intake of NMN may effectively enhance NAD+ levels, potentially preventing age-related muscle dysfunctions in humans.[10,1]

Since most studies start with cellular or animal levels as their sample subjects, researchers also confirmed that giving NMN supplementation can increase the NAD levels in mice. Although not directly in the blood, a study by Ramanathan *et al*. shows an increase in NAD levels in mice's brains with NMN supplementation within the 300-500 mg/kg range.[18] Another example from a study by Mills et al. noted a marginal elevation in plasma NAD+ levels can rise as swiftly as 2.5 minutes and restore at 15 minutes after giving 300 mg/kg of NMN oral supplementation. Nevertheless, the distinction did not reach statistical significance compared to their control group.[19] These findings suggest that NMN can be rapidly assimilated, effectively transported in the bloodstream, and absorbed and converted into NAD+ across various tissues. Augmenting NAD+ biosynthesis using NMN could be a practical therapeutic approach for various disease conditions.[18,19]

Introducing NMN into the body can restore the reduction in NAD+ levels caused by NAD+-consuming enzymes, as NMN serves as an intermediate compound in the biosynthesis of NAD+.[20] NMN serves as an intermediary by-product within the salvage pathway, playing a role in the synthesis of NAD+ through both salvage and Preiss-Handler pathways. The most effective and primary route for the biosynthesis of NAD+ is the salvage pathway. During this procedure, nicotinamide and 5-phosphoribosyl-1-pyrophosphate undergo conversion into NMN facilitated by the enzymatic activity of NAMPT. Following that, ATP and NMN combine, transforming into NAD through the action of nicotinic acid mononucleotide adenylyltransferase (NMNAT).[21]

Moreover, enzymes that consume NAD+ play a role in breaking down NAD+ and generating nicotinamide as a resulting by-product. Within the Preiss-Handler pathway, the first stage entails transforming nicotinic acid into nicotinic acid mononucleotide (NAMN) with the involvement of the nicotinic acid phosphoribosyl-transferase enzyme (NAPRT). After this, the nicotinic acid adenine dinucleotide (NAAD+) synthesis occurs, converting NAMN into NAAD+ through NMNAT 1/2/3. Then, NAAD+ is transformed into NAD+ by NAD+ synthetase (NADS) using ATP and ammonia.[22]

Presently, numerous studies indicate that NMN, the product of the NAMPT reaction, can be employed to stimulate SIRT1 activity. In instances where NAD+ levels are insufficient, SIRT1's capacity to impede hypoxia-inducible factor 1 alpha (HIF-1) diminishes, leading to elevated levels that disrupt communication not only at the cellular level between mitochondria and the nucleus but also at the systemic level between adipose tissue and the hypothalamus.[23] The consequent disruption in communication between mitochondria and the nucleus results in a rapid decline in mitochondrial function, contributing to age-related complications and diseases. However, administering NMN as a precursor to NAD+ can reinstate specific communication pathways and improve mitochondrial function.[21]

There are some limitations in this present study. The difficulty of access to contact the researchers of each study reviewed, secondary data sources obtained from the results of previous studies are not original research data that is open to the public, making it difficult to detect bias in data analysis. In addition, researchers fulfil the critical appraisal process using the JBI checklist, which allows bias because the assessment may be subjective. As an additional suggestion, this study only used a placebo as a comparison, for future studies expected to use studies with other comparisons. While this systematic review confirmed a noteworthy and satisfactory elevation in NAD+ levels in whole blood, further evaluations with a more extensive sample size are necessary to confirm the associations between individual factors and the elevated levels of NAD+ after taking NMN. Other research is warranted to investigate the impact of NMN supplementation on additional outcomes, for example, the efficacy of NMN supplementation from the capacity of healthy adult's endurance.

CONCLUSION

Our systematic review concludes that NMN supplementation significantly improves blood NAD concentration using varied doses starting from 250-900 mg/day, lasting between 12 weeks and 60 days, without serious adverse events. More multicenter randomized control conducting further studies is essential to obtain more reliable and clear results with minimal bias concerning the impact of NMN supplementation on anti-aging in adults.

Conflict of Interests

We declare no conflicts of interest in this study.

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