J Neural Transm (2000) 107: 1475–1481

Journal of Neural Transmission

No evidence for cognitive improvement from oral nicotinamide adenine dinucleotide (NADH) in dementia

Short Communication

M. Rainer[1], **E. Kraxberger**[1], **M. Haushofer**[1], **H. A. M. Mucke**[2], and **K. A. Jellinger**[3]

[1] Memory-Clinic and Psychiatric Department, Donauspital, Sozialmedizinisches Zentrum Ost,
[2] H.M. Pharma Consultancy, and
[3] Ludwig Boltzmann-Institute for Clinical Neurobiology, Vienna, Austria

Received March 27, 2000; accepted September 20, 2000

Summary. Reduced nicotinamide adenine dinucleotide (NADH) is advertised as an over-the-counter product or dietary supplement to treat Alzheimer's disease. We performed a 3-month open-label study with oral 10 mg/day NADH with 25 patients with mild to moderate dementia of the Alzheimer, vascular, and fronto-temporal types in addition to their current cholinomimetic drug medication. In 19 patients who completed the study, we found no evidence for any cognitive effect as defined by established psychometric tests. We conclude that NADH is unlikely to achieve cognitive improvements in an extent reported earlier, and present theoretical arguments against an effectiveness of this compound in dementia disorders.

Keywords: Alzheimer disease, dementia, nicotinamide adenosine dinucleotide, blood brain barrier, open clinical trial.

Introduction

Primary degenerative dementia of the Alzheimer type continues to puzzle clinicians and neurobiologists because the cause of neurodegeneration and the nature of the underlying degenerative process continue to be elusive. While the cholinergic system is most extensively affected in patients with Alzheimer's disease (AD), monoaminergic and other neuromodulator systems also show less severe deficits (Gsell et al., 1996). Increasing evidence demonstrates that oxidative stress and mitochondrial dysfunction largely contribute to neural damage in AD (Markesberry, 1999; Casserino and Bennett, 1999). Therefore, the use of antioxidants and/or free radical scavengers alone or in combination with cholinomimetic agents has been proposed

for both therapy and prevention of primary degenerative dementia (Pitchumoni and Doraiswamy, 1998; Prasad et al., 2000). Indeed, several free radical scavengers such as vitamin E with and without selegiline (Sano et al., 1997), and *Ginkgo biloba* extract EGb 761 (La Bars et al., 1997) have produced statistically significant positive results in controlled large-scale clinical trials. Cognitive effects have, however, been limited in these trials, with improvements that are lower than those obtained with acetylcholinesterase inhibitors that can achieve a relative benefit of 2–3 points on the Mini-Mental State Exam (MMSE) scale (Folstein et al., 1997), or 3–5 points on the cognitive subscale of the Alzheimer Disease Assessment Scale (ADAS-Cog.) over trial periods of three months, and a 6 to 12 months delay in symptomatic decline (Francis et al., 1999; Gauthier, 1999).

In contrast to this broad evidence, Birkmayer (1996) reported MMSE improvements of 6–14 (mean 8.35) points and 1–2 (mean 1.82) points in the Global Deterioration Scale (GDS) over 8–12 weeks in an open-label pilot study in 17 patients with AD, aged 33–84 (mean 67.7) years who received oral doses of reduced nicotine adenine dinucleotide (NADH; 10 mg b.i.d.). In addition to these impressive cognitive effects, all patients were reported to have improved 1 or 2 points on the Global Deterioration Scale (GDS), which is also in contrast to generally accepted evidence. This NADH tablet product has been extensively promoted as a dietary supplement claiming efficacy in AD and Parkinson's disease (PD) as well as in chronic fatigue syndrome (NADH, 1999; Birkmayer, 1999; web site http://www.enada.com and others; Physicians CFS study abstract, 1999). We have therefore attempted to replicate these findings in a cohort of demented subjects using a similar design.

Material and methods

Between March and November 1999 25 patients with mild or moderate probable AD according to the NINCDS-ADRDA (McKhann et al., 1984) criteria or vascular dementia according to the NICDS-AIREN (Roman et al., 1993) criteria and one case of frontotemporal dementia were enrolled in an open-label outpatient study that measured GDS and ratings on the MMSE as well as on the cognitive subscale of the AD Assessment Scale (ADAS-Cog). They were clients of a memory clinic managed by one of us (M. R.). The treatment schedule of 10 mg NADH (ENADA™, 2 × 5 mg tablets) given daily 30 min. before breakfast, corresponded to the regimen used by Birkmayer (1996). Since ENADA is a registered OTC product in Austria, no ethics committee approval was required for this study.

Psychometric measurements were performed at baseline, at 5–6 weeks, and at 10–12 weeks by a certified psychologist who was blinded to the treatment. Four patients were not evaluable because they did not present for the follow-up examinations; two were excluded because they discontinued their pre-existing acetylcholinesterase inhibitor medication, and experienced significant cognitive withdrawal effects. Complete data were available for the remaining 19 patients (10 male, 9 female; age 54–91 years). Their median baseline scores on the GDS, MMSE ADAS-Cog scales were 4 (range 3–5), 17 (10–26) and 34 (12–57), respectively, corresponding to a moderate cognitive deficit.

Results

All patients completed the study course without reporting any adverse events conceivably related to NADH. No clinically relevant changes versus baseline

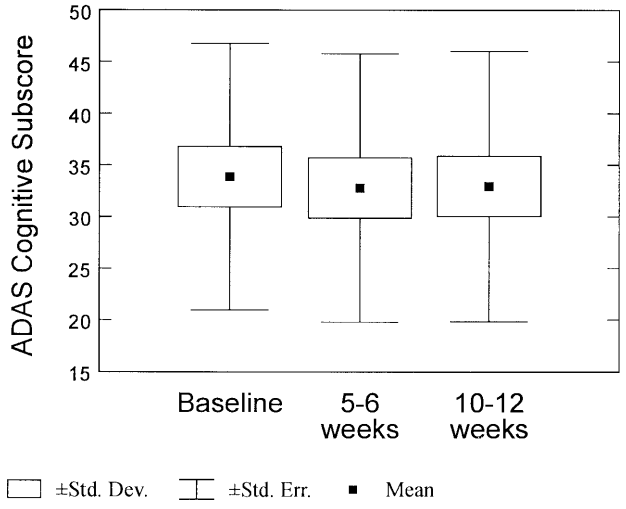

Fig. 1. ADAS Cognitive Subscore after NADH treatment

were seen in the GDS, the cognitive parameters, or any of the three subscores of the ADAS-Cog that capture memory, orientation and language in the group as a whole (Fig. 1). Moreover, none of the minute changes that where observed in these parameters achieved statistical significance ($p < 0.01$) or indicated a statistical trend ($p < 0.05$) when the t-test for dependent samples was applied (Table 1).

As expected, broad individual variation was seen among the 19 individuals who completed the study. The MMSE score differences varied from −5 to +8

Table 1. Changes of global and cognitive parameters versus baseline at midpoint and endpoint. Note that, while MMSE scores decrease with a decrease in cognition, GDS and ADAS-Cog scores are inversely related to global and cognitive patient status, respectively

	Differential at 5–6 weeks	Differential at 10–12 weeks	Means (SD)		
			baseline	5–6 weeks	10–12 weeks
Global Deterioration Score (GDS)	0.10	0.19	4.37 (0.60)	4.47 (0.80)	4.56 (1.04)
Mini-Mental State Exam Score (MMSE)	0.10	0.58	16.95 (5.09)	17.05 (5.53)	17.53 (5.76)
Alzheimer Disease Assessment Scale Cog. Subscore (ADAS-Cog)	−1.21	−1.16	33.95 (12.75)	32.74 (12.87)	32.79 (13.03)
ADAS-Cog. Subscore Memory	0.84	1.31	12.95 (4.90)	13.79 (5.35)	14.26 (4.89)
ADAS-Cog. Subscore Orientation	−0.66	−1.08	11.61 (4.92)	10.95 (5.41)	10.53 (6.54)
ADAS-Cog. Subscore Language	−1.38	−1.75	7.33 (2.79)	5.95 (3.60)	5.58 (3.49)

points and the ADAS-Cog from +10 to −9 points, corresponding to the entire range from deterioration in one dimension to improvement in two dimensions. There was no significant correlation between this outcome after 3 months and the respective baseline values, or with the subtype classification of the Alzheimer component of dementia (2 cases early-onset, ICD-10 classification F00.0; 10 cases late-onset, F00.1; 7 mixed-type cases, F00.2).

Discussion

Using an open-label design similar to that employed in the original study, and a comparable number of patients who attended a memory clinic, we were unable to replicate earlier results indicating a dramatic effect of oral NADH application in dementia patients (Birkmayer, 1996).

What is the supposed rationale for NADH supplementation in AD? Pathological alterations of mitochondrial cytochrome c oxidase, which relies on the NAD/NADH redox pair for functioning, have been described in thrombocytes (Zubenko, 1989) and in the brains of AD patients (Zubenko et al., 1990). It has also been known for more than a decade that the activity of the enzyme NADH oxidoreductase (formerly known as diaphorase), which is coupled to the mitochondrial respiratory chain, is increased in both the neocortex and the hippocampus of AD patients, most likely in association with amyloid plaques, one of the hallmarks of AD dementia (Jacobs et al., 1985; Nakamura et al., 1987). Another NAD/NADH-dependent hydroxyacyl-coenzyme A dehydrogenase in human brain (He et al., 1998) has recently been found to be identical with a β-amyloid peptide-binding protein (ERAB) that might mediate some of the neurotoxic effects of amyloid (Yan et al., 1997).

While these data may be considered to provide a promising starting point for the application of NADH in AD, Birkmayer (1996) rather argued that the rationale for his study was the efficacy of NADH in PD that has been repeatedly reported (Birkmayer et al., 1989, 1993; Birkmayer and Birkmayer, 1989; Kuhn et al., 1996). The biochemical basis of this therapy is that NADH can stimulate the endogenous synthesis of dopamine by increasing the activity of quinonoid dihydropteridine reductase, the enzyme which recycles the inactive dihydrobiopterin to the active tetrahydrobiopterin, thereby providing reduction equivalents to tyrosine hydroxylase (Vrecko et al., 1997), the rate-limiting factor of dopamine biosynthesis. However, diminished dopamine synthesis – while probably responsible for some aspects of depression in AD (Gsell et al., 1996) – is not known to play a significant role in its typical cognitive deficits. In addition, other suggested favourable effects of NADH on cell repair and on the immune system have never been confirmed (Jellinger, 1999).

Moreover, the central question of cerebral bioavailability of NADH has not been addressed so far, and results on the pharmacology of NADH are not available from peer-reviewed literature, except for cursory remarks that preclinical safety studies with 500 mg/kg/day NADH i.v. produced no deaths in beagle dogs during 14 days (Birkmayer, 1996). Biochemical textbook

knowledge states that NADH, unlike ascorbic acid which is actively taken up by the brain in the oxidized state (Agus et al., 1997), does not penetrate the blood brain barrier under physiological concentrations (Swerdlow, 1998). Unless the existence of a facultative transport mechanism is demonstrated that could shuttle either NADH or reduction equivalents between the separate NAD/NADH pools in the central and peripheral compartment, there is no basis for the assumption that the administration of exogenous NADH may have any direct effect on brain metabolism. By contrast, it is conceivable that high doses (certainly much more than 10 mg/day used in available studies) of externally administered NADH might inhibit the acquisition of ascorbate by the brain by inhibiting its oxidation to dehydroascorbate.

Finally, to the best of our knowledge, no double blind, placebo controlled study in patients with AD has been reported so far, although the results of an ongoing study in the USA were announced to be available by the end of 1998 (NADH, 1999). While our study design did not have sufficient statistical power to demonstrate the existence or absence of a cognitive effect of oral NADH treatment in a clinically relevant dimension, it certainly would have been able to replicate the dramatic improvement reported in the first open study (Birkmayer, 1996). We rather concur with Birkmayer's (1995) remark in his pertinent patent where he states this finding to be "surprising and completely unexpected". Considering the clinical evidence, and the theoretical considerations that are outlined above and have already been emphasized (Jellinger, 1999), at present we cannot recommend the use of NADH to improve cognitive impairment in AD, until controlled clinical trials have unequivocally demonstrated its efficacy.

References

Agus DB, Gambhir SS, Pardridge WM, Spielholz C, Baselga J, Vera JC, Golde DW (1997) Vitamin C crosses the blood-brain barrier in the oxidized form through the glucose transporters. J Clin Invest 100: 2842–2848

Birkmayer GJ, Birkmayer W (1989) Stimulation of endogenous L-dopa biosynthesis – a new principle for the therapy of Parkinson's disease. The clinical effect of nicotinamide adenine dinucleotide (NADH) and nicotinamide adenine dinucleotidephosphate (NADPH). Acta Neurol Scand [Suppl] 126: 183–187

Birkmayer JG (1995) Method for treating symptoms of Alzheimer's with NADH and NADPH. United States Patent No. 5, 444, 053; August 22, 1995

Birkmayer JG (1996) Coenzyme nicotinamide adenine dinucleotide: new therapeutic approach for improving dementia of the Alzheimer type. Ann Clin Lab Sci 26: 1–9

Birkmayer JG (1999) NADH, the energizing coenzyme. How an important, yet little-known coenzyme enhances cellular energy in the brain and body functions. Keats Publishing, Chicago

Birkmayer JG, Vrecko C, Volc D, Birkmayer W (1993) Nicotinamide adenine dinucleotide (NADH) – a new therapeutic approach to Parkinson's disease. Comparison of oral and parenteral application. Acta Neurol Scand [Suppl] 146: 32–35

Birkmayer W, Birkmayer GJ, Vrecko K, Mlekusch W, Paletta B, Ott E (1989) The coenzyme nicotinamide adenine dinucleotide (NADH) improves the disability of parkinsonian patients. J Neural Transm [PD-Sect] 1: 297–302

Cassarino DS, Bennett JP (1999) An evaluation of the role of mitochondria in neurodegenerative diseases: mitochondrial mutations and oxidative pathology, protective nuclear responses, and cell death in neurodegeneration. Brain Res Brain Res Rev 29: 1–25

Folstein M, Folstein SE, McHugh PR (1975) "Mini Mental State": a practical method for grading the cognitive state of patients for the clinician. J Psychiatr Res 12: 189–198

Francis PT, Palmer AM, Snape M, Wilcock GW (1999) The cholinergic hypothesis of Alzheimer's disease; a review of progress. J Neurol Neurosurg Psychiatry 66: 137–147

Gauthier S (ed) (1999) Clinical diagnosis and management of Alzheimer's disease, 2nd ed. Martin Dunitz, London

Gsell W, Strein I, Riederer P (1996) The neurochemistry of Alzheimer type, vascular type and mixed type dementias compared. J Neural Transm [Suppl 47]: 73–101

He XY, Schulz H, Yang SY (1998) A human brain L-3-hydroxyacyl-coenzyme A dehydrogenase is identical to an amyloid beta-peptide-binding protein in Alzheimer's disease. J Biol Chem 273: 10741–10746

Jacobs RW, Farivar N, Butcher LL (1985) Alzheimer dementia and reduced nicotinamide adenine dinucleotide (NADH)-diaphorase activity in senile plaques and the basal forebrain. Neurosci Lett 53: 39–44

Jellinger KA (1999) NADH (Nikotinsäure-Adenin-Dinukleotid) und M. Alzheimer. Literatur-Recherche und kritische Bemerkungen. Neuropsychiatrie 13: 85–87

Kuhn W, Muller T, Winkel R, Danielczik S, Gerstner A, Hacker R, Mattern C, Przuntek H (1996) Parenteral application of NADH in Parkinson's disease: clinical improvement partially due to stimulation of endogenous levodopa biosynthesis. J Neural Transm 103: 1187–1193

Le Bars PL, Katz MM, Berman N, Itil TM, Freedman AM, Schatzberg AF (1997) A placebo-controlled, double-blind, randomized trial of an extract of Ginkgo biloba for dementia. North American EGb Study Group. J Am Med Asscoc 278: 1327–1332

Markesberry WR (1999) The role of oxidative stress in Alzheimer disease. Arch Neurol 56: 1449–1452

McKhann G, Drachman D, Folstein M, Katzman R, Price D, Stadlan EM (1984) Clinical diagnosis of Alzheimer's disease: report of the NINCDS-ADRDA Work Group under the auspices of Department of Health and Human Services Task Force on Alzheimer's disease. Neurology 34: 939–944

NADH (1999) Alzheimer's disease (part 1 to 3) mod. version 18.3.1999. Renascent Systems Inc

Nakamura S, Kawamata T, Akiguchi I, Kameyama M, Kimura H (1987) Reduced nicotinamide adenine dinucleotide phosphate diaphorase histochemistry in neocortex and hippocampus in patients with Alzheimer type dementia and aged controls. Rinsho Shinkeigaku 27: 1059–1063

Physician CFS study abstract (1999) NADH a new therapeutic approach in chronic fatigue syndrome (CFS). MENUCO Corp

Pitchumoni SS, Doraiswamy PM (1998) Current status of antioxidant therapy for Alzheimer's disease. J Am Geriatr Soc 46: 1566–1572

Prasad KN, Hovland AR, Cole WC, Prasad KC, Nahreini P, Edwards-Prasad J, Andreatta CP (2000) Multiple antioxidants in the prevention and treatment of Alzheimer disease: analysis of biologic rationale. Clin Neuropharmacol 23: 2–13

Román GC, Tatemichi TK, Erkinjuntti T, et al (1993) Vascular dementia: diagnostic criteria for research studies. Report of the NINDS-AIREN International Workshop. Neurology 43: 250–260

Sano M, Ernesto CH, Thomas RG, Klauber MR, Schafer K, Grundman M, Woodbury P, Growdon J, Cotman CW, Pfeiffer R, Schneider LS (1997) A controlled trial of selegiline, alpha-tocopherol, or both as treatment for Alzheimer's disease. N Engl J Med 336: 1216–1222

Swerdlow RH (1998) Is NADH effective in the treatment of Parkinson's disease? Drugs Aging 13: 263–268

Vrecko K, Storga D, Birkmayer JG, Moller R, Tafeit E, Horejsi R, Reibnegger G (1997) NADH stimulates endogenous dopamine biosynthesis by enhancing the recycling of tetrahydrobiopterin in rat phaeochromocytoma cells. Biochim Biophys Acta 1361: 59–65

Web sites http://www.enada.com and others

Yan SD, Soto C, Chen X, Zhu H, al Mohanna F, Collison K, Zhu A, Stern E, Saido T, Tohyama M, Ogawa S, Roher A, Stern D (1997) An intracellular protein that binds amyloid-beta peptide and mediates neurotoxicity in Alzheimer's disease. Nature 389: 689–695

Zubenko GS (1989) Endoplasmic reticulum abnormality in Alzheimer's disease: selective alteration in platelet NADH-cytochrome c reductase activity. J Geriatr Psychiatry Neurol 2: 3–10

Zubenko GS, Moossy J, Claassen D, Martinez AJ, Rao GR (1990) Brain regional analysis of NADH-cytochrome C reductase activity in Alzheimer's disease. J Neuropathol Exp Neurol 49: 206–214

Authors' address: Dr. M. Rainer, Memory-Clinic and Psychiatric Department, Donauspital, Sozialmedizinisches Zentrum Ost, Langobardenstrasse 122, A-1220 Wien, Austria.