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The Clincial Benefit of NADH as Stimulator of Endogenous L-Dopa Biosynthesis in Parkinsonian Patients

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Treatment of Parkinson's disease with L-dopa (LD) in combination with decarboxylase and monoamine oxidase inhibitors is a pure substitutional therapy designed to correct the lack of dopamine in the brain (1). The dopamine deficit is caused by the diminished tyrosine hydroxylase (TH) in the substantia nigra (2). However, catecholamines such as dopamine and its precursor LD inhibt TH via a feedback mechanism (3,4). This suggests that application of LD to PD patients may further decrease the already reduced TH activity. Therefore, therapeutic strategies other than substitution have to be considered, e.g., stimulation of endogenous dopamine production in the brain. This may be achieved by activating the LD producing enzyme.

As shown by Nagatsu et al. (5), this enzyme is an iron-containing protein with tetrahydrobiopterin (H4biopterin) as coenzyme. H4biopterin is reduced in the brain of PD patients (6) and has therefore been used in clincial trials, but with only partial success. On the other hand, it has been shown that the special iron compound oxyferriscorbone(R) is able to improve the symptoms of parkinsonian patients (7,8), suggesting production of endogenous LD in the brain of parkinsonians. The stimulation of LD biosynthesis is reflected by an increase in the urine level of homovanillic acid (HVA) (8). We believe that this occurs by TH activation by the iron compound oxyferriscorbone, because there is no other enzyme except TH which catalyzes LD formation, and it also has been shown that this enzyme can be markedly activated *in vitro* by iron (9). Our findings have already been confirmed (10,11). After long-term iron medication, however, its effectiveness subsides in some patients. This prompted us to look for other therapeutic modalities. Our choice was nicotinamidadenindinucleotide (NADH), which promotes the formation of H4biopterin, the active coenzyme of tyrosine hydroxylase.

MATERIALS AND METHODS

Diagnosis and disability scores of the parkinsonian patients were established according to the method of Birkmayer and Neumayr (12). Nicotinamideadeninedinucleotide, reduced form disodium salt (synonyms: betaNADH, reduced DPN, beta-DPNH), was purchased from Sigma Diagnostics (St. Louis, Missouri). Twenty-five mg of NADH were dissolved in 100 ml of 0.9% sterile sodium chloride, pH 7.4, and infused intravenously in 30 min. NADH solutions were always prepared fresh im-

mediately prior to use. Disability scores were determined before, 1 hr after, and 4 hr after the NADH infusion.

RESULTS

Forty patients have been treated so far. All of them exhibited a pronounced drop of their disability score. Table 1 summarizes all the details of the individual patients: their age, duration of disease, disability, therapy, and treatment duration.

The patients' ages ranged from 48 to 85, duration of the disease ranged from 2 to 20 years. The overall improvement of the disability of all patients was 46.25%. A very good response was exhibited in 65% of the patients: more than 30% improvement of disability, 35% of the patients a moderate benefit of up to 30% (Fig. 1).

TABLE 1. *Improvement of PD disability after NADH treatment*

Patient	Age	Duration (years)	Percent disability before NADH	Percent disability after NADH
(1) EN	65	12	60	10
(2) KO	70	19	70	40
(3) OR	56	4	50	20
(4) KO	62	8	60	45
(5) EV	72	10	70	20
(6) MA	66	12	70	20
(7) PR	74	14	50	30
(8) BL	66	11	80	40
(9) MA	56	6	70	20
(10) PR	66	14	75	25
(11) PA	58	3	40	25
(12) VA	70	8	30	15
(13) UL	40	2	30	20
(14) ST	70	6	50	40
(15) TO	71	8	60	30
(16) MI	61	15	70	30
(17) KE	70	8	75	20
(18) TR	56	8	60	20
(19) RE	62	8	75	25
(20) RO	62	8	50	20
(21) JA	62	6	70	20
(22) NA	85	15	80	40
(23) BE	72	20	75	40
(24) BE	62	12	70	20
(25) LA	48	4	40	20
(26) FU	72	15	70	20
(27) PL	71	10	75	40
(28) WE	70	10	70	20
(29) SC	63	10	60	20
(30) KO	70	19	70	20
(31) ST	84	12	80	50
(32) ST	65	10	60	30
(33) GR	75	15	50	15
(34) GR	66	16	60	20
(35) RK	48	2	40	10
(36) AD	54	4	60	20
(37) BA	58	5	50	25
(38) SB	62	6	60	20
(39) BS	59	4	40	20
(40) GW	58	3	40	10

PD, Parkinson's disease; NADH, nicotinamideadeninedinucleotide.

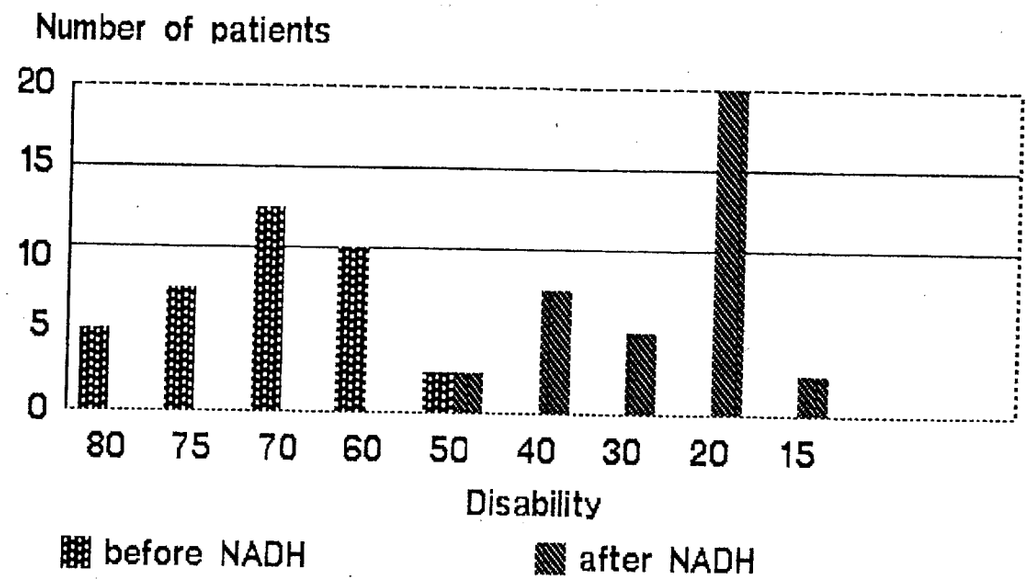

FIG. 1. Improvement of disability by NADH. The disability before NADH treatment was in the range of 80 to 50. After NADH treatment the disability dropped to 40 up to 15. The over all improvement in disability of all patients was 46.25%.

Walking and pushing ability improved considerably as did posture, speech, and mimics. The action of NADH lasted between 1 and 4 days, depending on the severity of the symptoms. Withdrawal of NADH lead to a relapse with worsening of disability. About one-fifth of the patients did well on NADH alone, and LD-therapy could be omitted. In the other patients the LD dosage could be reduced materially.

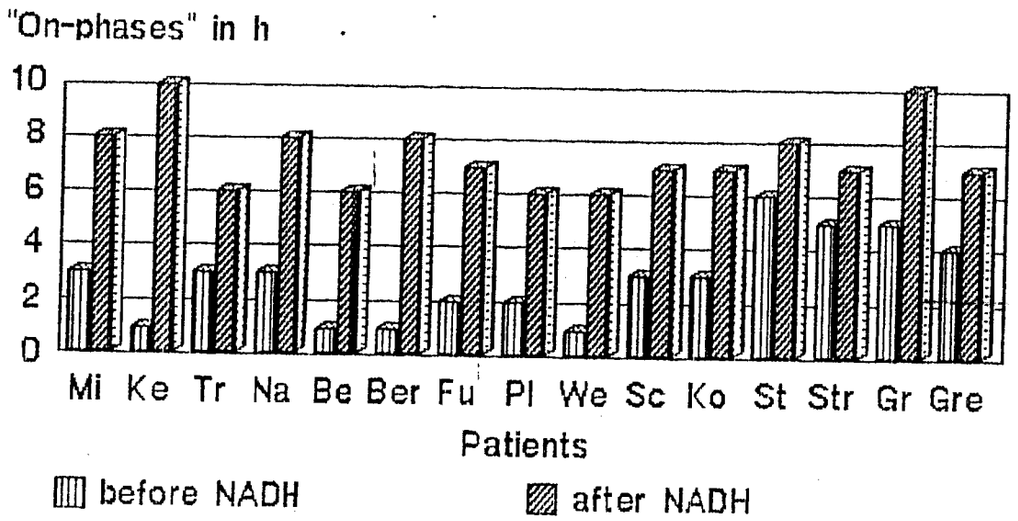

FIG. 2. Prolongation of "on-phases" by NADH. From the 15 patients examined with regard to the duration of the daily "on-phases," all showed a prolongation. This prolongation lasted from 2 hr to 10 hr in the individual patients.

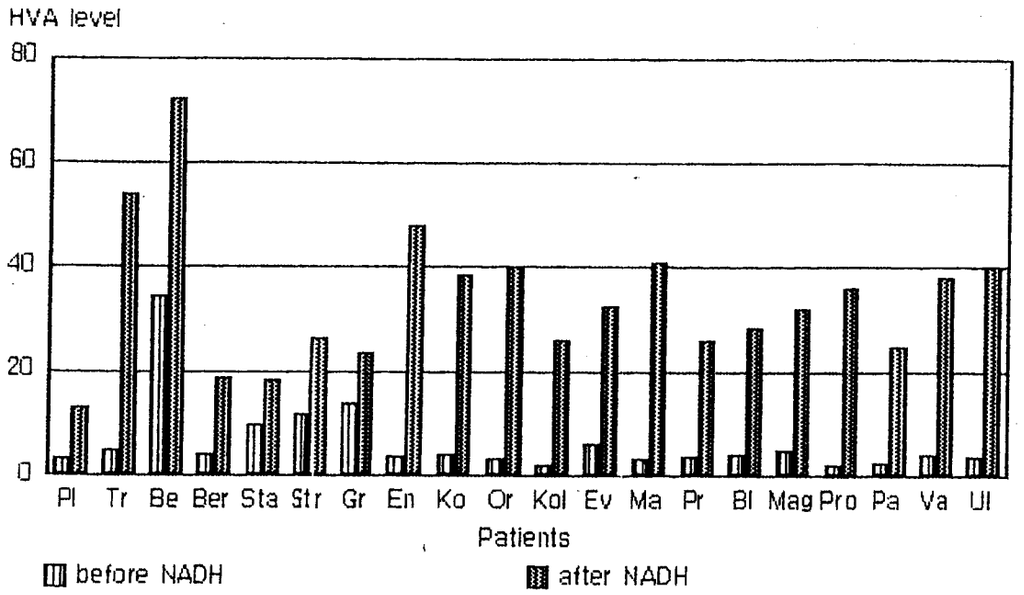

FIG. 3. HVA urine level after NADH treatment. In 20 patients who showed an improvement in disability, the homovanillic acid (HVA) urine concentration was determined. In all these 20 patients a considerable increase of the HVA urine level could be determined, indicating an increase in dopamine biosynthesis.

Fifteen patients have been examined with regard to the duration of the daily pattern of phases (Fig. 2). The daily "on" phases could be increased by 6 to 10 hr. In a number of patients so examined, the urine level of the dopamine metabolite HVA was markedly increased (Fig.3). Such HVA increase is also observed after treatment with LD. As NADH is not a precursor of LD, it seems most likely that it stimulates endogenous LD biosynthesis.

DISCUSSION

The beneficial clinical effect of NADH on the disability of parkinsonian patients has been demonstrated. A possible mechanism by which this may occur is shown in the simplified pathway of LD biosynthesis in Fig. 4. L-Dopa is formed from tyrosine by the enzyme TH. This is an iron-containing enzyme with H4biopterin as coenzyme. H4Biopterin provides electrons to reduce molecular oxygen and is in turn oxidized to the quinonoid-H2-pterin. The dihydropteridine reductase (DHPR) regenerates H4biopterin. The cofactor of this enzyme is NADH (13). H4Biopterin in brain and in cerebrospinal fluid of PD patients is reduced (6). This H4biopterin deficiency could be due either to a decreased biosynthesis or to a lack in the biologically active form of H4biopterin. It could well be that H4biopterin is exhausted in parkinsonian patients because of an enormous consumption, perhaps caused by a toxic agent. The idea is derived from the observation that MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine), a neurotoxin that can induce parkinsonism in men and animals, inhibits DHPR, the enzyme which regenerates tetrahydrobiopterin, the coenzyme required for endogenous dopamine formation (14). MPTP seems to be a competitive inhibitor

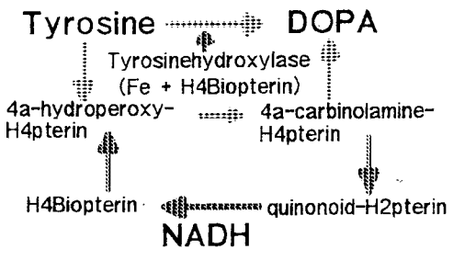

FIG. 4. Possible mechanism of NADH action. L-Dopa is formed from tyrosine via the enzyme tyrosine hydroxylase. This enzyme is the rate limiting step in the biosynthesis of L-dopa and is considerably reduced in the brain of parkinsonian patients. In addition the coenzyme of tyrosine hydroxylase, H4biopterin, is also considerably reduced. H4Biopterin is formed from H2pterin by an enzyme called dihydropteridine reductase (DHPR). The coenzyme of DHPR is NADH. Implying that NADH is an essential component for the regeneration of H4biopterin. This indicates that NADH is involved in the biosynthesis of H4biopterin and therefore in the activation of tyrosine hydroxylase.

of DHPR with respect to NADH. If this is so, NADH should be able to neutralize the toxic affect of MPTP or other free radical-inducing agents. There are two arguments in favor of our hypothesis. First, the clinical effect of NADH closely resembles that of LD, indicating that this coenzyme stimulates the endogenous LD biosynthesis. Second, the improvement in the clinical symptoms parallels an increase in the urine level of HVA, which also has been found with the standard LD therapy.

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