J Neural Transm (2000) 107: 815–829

Journal of Neural Transmission

Sustained improvements in patients with dementia of Alzheimer's type (DAT) 6 months after termination of Cerebrolysin therapy

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Received August 4, 1999; accepted November 24, 1999

Summary. The present study is an extension of the work of Rüther et al. (1994). 101 patients suffering from DAT were evaluated 6 months after completion of a 4 week (5 days per week) therapy with either 30 ml Cerebrolysin or placebo. The significant and clinically relevant improvements in the global rating (CGI), clinical symptomatology (SCAG), cognitive performance (ZVT-G) as primary efficacy variables, as well as the improvements in the secondary efficacy variables activities of daily living (NAI) and wellbeing (Bf-S), achieved in the Cerebrolysin group after only 4 weeks of active therapy, were maintained to a large extent during the follow-up period. Although there was a moderate tendency in the drug group towards loss of improvement, the differences between baseline and follow-up examination, as well as the differences between the verum and the placebo group, clearly document a sustained improvement in patients treated with Cerebrolysin in the first 4 weeks of the study period. It can be speculated that relatively short treatment courses with Cerebrolysin in patients suffering from neurodegenerative dementia can lead to long term influence on disease progression, which is in accordance with the proposed neurotrophic – nerve growth factor like – mode of action.

Keywords: Alzheimer's disease, Cerebrolysin, neurotrophic therapy, disease progression.

Introduction

In the western industrial countries dementia of Alzheimer's type is the 4th most common cause of death, behind heart disease, cancer, and stroke (Bush et al., 1994). In conjunction with increasing life expectancy, the exponential

increase in prevalence between the 6th (0.07%) and 9th (38.6%) decades of life will lead to a doubling in case numbers of Alzheimer's patients within the next 40 years (Evans, 1990; Keefover, 1996). In addition to the preservation and/or improvement of cognitive performance, the most important goal of pharmacological treatment is the maintenance or improvement of the activities of daily living, to prolong the time period of functional independence.

The first therapeutic attempts for Alzheimer disease were focussed on the deficit of central cholinergic neurotransmission which is associated with disease symptoms (Patel, 1995). Direct cholinergic replacement strategies turned out not to be successful, but the use of acetylcholinesterase inhibitors resulted in an improvement of the symptoms of mild to moderate DAT (Taylor, 1998; Farlow and Evans, 1998). The first approved cholinesterase inhibitor was Tacrine, showing modest, but reproducible improvements in cognitive performance, in most studies documented by the Alzheimer disease assessment scale cognitive subscale (ADAS-Cog) and treatment also resulted in an improvement of the global rating scales (Eager et al., 1991; Davis et al., 1992). Its use was just limited by its unfavorable side effect profile with signs of hepatotoxicity in a high proportion of patients (Knapp et al., 1994). However in the meanwhile two other substances are available in most countries. Donepezil (Shintani, 1997; Rogers, 1998) displays a very good tolerability and has the advantage of a daily single oral dosing. Rivastigmine (Anand, 1996; Rösler, 1999) shows more cholinergic side effects but only of mild to moderate degree. Both of this second generation cholinesterase inhibitors produce reproducible improvements of cognitive performance, global functioning and in some of the studies also of activities of daily living. They share these activities with other cholinesterase inhibitors under development like the long acting drug Metrifonate (Williams, 1999) or Galanthamine (Wilcock, 1997). It is of interest that the achieved improvements with cholinesterase inhibitors are in the same range independent from the used drug, in spite of considerable differences in the mode and strength of enzyme inhibition or the selectivity for acetylcholinesterase versus butyrylcholinesterase. Hopes that a cholinergic strategy can lead to a disease modification, due to an influence on amyloid precursor protein processing (Younkin, 1998) are not fulfilled, as shown in studies with drug withdrawal after half a year of treatment (Rogers, 1998) which was leading to a rapid deterioration in cognitive performance, down to the levels of placebo-treated controls.

Other treatment strategies, like the use of the MAO-B inhibitor Selegeline (Wessel, 1993; Freedman et al., 1998) or the use of nootropic drugs like Piracetam (Croisile et al., 1993), Idebenone (Gutzman and Hadler, 1998; Weyer et al., 1996), standardized Ginkgo biloba extracts (Maurer et al., 1998) or the use of gangliosides (Svennerholm, 1994) brought positive effects, but did not cumulate enough data until now to get approval for the treatment of symptoms of Alzheimer's disease from health authorities.

A substantial hope was put into the use of naturally occurring neurotrophic factors, like nerve growth factor (NGF), especially since the discovery that the substance is selectively acting on central nervous system cholinergic neurons (Johannsen, 1988; Thönen et al., 1988). The strongly

advocated therapeutic use for treatment of Alzheimer's disease (Sofroniev, 1996) resulted in some disappointing results from first limited clinical trials in few subjects (Olson et al., 1992; Seiger et al., 1993). Due to side effects like hyperalgesia and weight loss the treatment had to be terminated in two out of three patients. No improvements on cognition could be shown in the remaining subject completing the infusion period. However there were improvements in cerebral blood flow and EEG showed a reduction of slow waves which persisted almost one year after stop of treatment. Improvements of cerebral blood flow due to NGF treatment was also demonstrated by Nordberg (1997) and Jönhagen (1998). At the moment no further large scale clinical trials with neurotrophic substances are in progress, but there are ongoing scientific efforts to improve neurotrophic factor therapy for neurodegenerative disorders, by overcoming their problems with blood-brain barrier penetration and also creating substances acting on different growth factor receptors, so called pan-neurotrophins (Ibanez, 1993; Skaper, 1998).

The drug Cerebrolysin, a brain-derived peptide preparation produced by biotechnological methods using a standardized enzymatic breakdown of purified porcine proteins might be another approach towards neurotrophic treatment of DAT. This preparation is in use for treatment of dementia of different etiology and sequel of stroke or brain trauma in several European and Asian countries since many years. For a long time it was already known that Cerebrolysin is able to influence brain oxidative metabolism, exhibiting strongest effects in brains of old aged animals (Windisch et al., 1985). The also described significant influence on overall brain protein synthesis (Piswanger et al., 1990), in accordance with data published by Satou (1993, 1994) reporting that Cerebrolysin has nerve growth factor-like activities on chicken dorsal root ganglia. In control experiments using antibodies against naturally occurring NGF he could not block Cerebrolysin's activity, indicating that the active compounds are different from NGF. The neurotrophic activity of Cerebrolysin's low molecular weight peptides was shown in many other experiments also on tissue cultures of cortical neurons (Hutter-Paier, 1996a, 1998a; Windisch, 1998). Addition of the drug to the tissue culture medium resulted in enhanced sprouting of nerve fibers and it also prevented neuronal degeneration after different lesions, like serum withdrawal which induces apoptosis, glutamate intoxication or oxidative stress (Hutter-Paier, 1996b, 1998b). After unilateral transection of fimbria fornix, which induces degeneration of cholinergic neurons in the medial septum, a standard model to prove the biological activity of NGF in vivo, it was demonstrated that intraperitoneal injection of Cerebrolysin is able to rescue approximately 60% of the afflicted cholinergic neurons (Akai et al., 1992). Compared to NGF the drug was less effective, but it must be considered that the protection was achieved after peripheral injection, instead of the intracerebroventricular infusion which is necessary for application of NGF. This indicates additionally that at least a small proportion of Cerebrolysin's active molecules is able to penetrate blood-brain barrier in pharmacodynamically relevant amounts. This was also shown by an experiment comparing intracerebroventricular versus intraperitoneal injection of Cerebrolysin (Gschanes, 1997). After a fimbria fornix

transection animals develop pronounced memory and learning deficits. The treatment with Cerebrolysin improved the cognitive performance of rats after the fimbria fornix transection in a spatial orientation task using the Morris Water Maze up to the level of healthy controls. In this task it was more effective than NGF, and in contrast to this naturally occurring growth factor it improved not only memory but also acquisition of new information (Francis et al., 1996; Valouskova, 1998). In apolipoprotein E knock out mice an early neuronal degeneration occurs, mainly afflicting the dendritic region of the nerve cells. This degeneration is accompanied by disturbances in learning and memory. The chronic treatment of APOE-knock out mice for 4 weeks re-establishes normal microtubuli associated protein levels in frontal cortex, increases the synaptic density and exhibits morphological effects which are of suggestive a normalization of neuronal cytoarchitecture. Also their cognitive deficit was significantly improved (Masliah et al., 1999). Such significant improvements of spatial learning and memory were also achieved after 4 weeks treatment of 24-month-old rats (Gschanes et al., 1998), which was associated with significant increases in synaptic density in the hippocampus and the entorhinal cortex (Reinprecht et al., 1999). Additional direct effects of Cerebrolysin on expression of blood-brain barrier glucose transporter, which are associated with an increased cerebral glucose transport (Boado et al., 1999) might improve the energy supplementation of the neurons. Very recent data (Lombardi et al., 1999) also suggest that the drug inhibits pathological microglia activation and the enhanced release of cytokines like Interleukin-1β (Lombardi et al., 1999).

For detailed review of Cerebrolysin's preclinical and clinical effects see Windisch et al. (1998).

Also a series of clinical studies with the drug Cerebrolysin (Hebenstreit, 1996; Suchanek-Fröhlich and Wunderlich, 1986; Kofler et al., 1989, 1990; Vereshagin et al., 1991) showed a therapeutic influence on cognitive function in patients suffering from dementia of different ethiology. These studies were performed with different dosages of Cerebrolysin ranging from 10 to 30 ml, already indicating a correlation between dosage and effect. In spite of the reported therapeutical effects conclusions from these studies must be interpreted with caution, because of small patient numbers.

More detailed information concerning the clinical effectiveness of Cerebrolysin therapy was provided by the results of a prospective, placebo controlled, double blind study of 120 Alzheimer's patients with mild to moderate symptoms (Rüther et al., 1994). Intravenous treatment with 30 ml Cerebrolysin daily for 4 weeks (5 days a week) led to significant improvements compared to the placebo group in all three primary efficacy variables: global rating (CGI), clinical symptoms (SCAG) and cognitive performance (ZVT-G). There was also a significant improvement of patient's self rated wellbeing (Bf-S) and activities of daily living (NAI).

The goal of the present follow-up study was to document a sustained improvement in these patients (Rüther et al., 1994) 6 months after end of active drug therapy. The primary (CGI, SCAG, ZVT-G) and secondary (Bf-S, NAI) efficacy criteria of this study are the same as in the previous study.

Material and methods

Following the clinical study of Rüther et al. (1994), in which patients were treated with 30 ml Cerebrolysin in 100 ml physiological saline or placebo for 4 weeks (5 days per week), a new evaluation of the psychopathological symptoms (SCAG) and cognitive performance in the trail making test was conducted. In addition, the activities of daily living (NAI, Oswald and Fleischmann, 1986), self assessment according to von Zerssen and Möller (1980) (Bf-S) and the Clinical Global Impressions (CGI) were evaluated. This re-evaluation was performed 6 months after end of the active treatment course, corresponding to 7 months after the baseline evaluation.

41 male and 79 female patients between 55 and 85 years suffering from mild to moderate senile dementia of Alzheimer's type (SDAT) according to DSM-III-R criteria were included in the study. A CT and an evaluation with the Hachinski Ischemic Score (Hachinski et al., 1975) were carried out to exclude secondary forms of dementia. At Baseline the Reisberg Global Deterioration Scale (GDS – Reisberg et al., 1982) and the Mini Mental State Examination (MMSE – Folstein et al., 1975) were used to assess the severity of the disease.

The following criteria were used to exclude patients from the study: transient organic psychoses; brain disease with focal symptoms; stroke; neurological deficits interfering with performance in the tests; status epilepticus; polyneuropathy; thyroid or parathyroid dysfunctions; untreatable hypertension; congestive heart failure; life-threatening diseases; allergic diathesis or drug allergy; situations unfavorable to proper test performance; existing drug or alcohol abuse; concomitant therapy with other nootropics; chronic consumption of nootropics, psychotropic drugs, hypnotics, stimulants, drugs influencing cerebral blood flow; participation in another clinical trial, refusal to take part in the trial. Detailed information on demographics, inclusion and exclusion criteria are provided by Rüther et al. (1994).

Cerebrolysin is a brain derived peptide preparation produced by a standardized enzymatic breakdown of lipid free proteins. It consists of low molecular weight peptides (nlt. 15% based on the total nitrogen content) stabilized with amino acids.

Although a statistical analysis was done after end of therapy (Rüther et al., 1994), this follow-up after another 6 months remained blinded, because the investigators did not get the information which patient belonged to placebo or verum groups.

Statistical analysis between the treatment and control groups was accomplished using the Wilcoxon-Mann-Whitney U-Test. The Signed Rank Test was used for comparison within each group. A level of significance of $p < 0.05$ was required for all tests.

Results

For the follow-up evaluation 101 patients were available 6 months after the end of the treatment period, corresponding to 84.2% of the subjects originally enrolled into the study of Rüther et al. (1994). Of them, 49 had been treated with Cerebrolysin, 52 with placebo. About one fourth of the patients had received additional therapy in the intervening 6 months. Thus 18 Cerebrolysin patients received additional therapy and 31 did not. In the placebo group 37 out of 52 patients received no further treatment (Table 1).

In the original study, significant ($p < 0.05$) improvements in the primary (CGI, SCAG, and ZVT-G) and secondary (NAI and Bf-S) efficacy criteria were achieved in the Cerebrolysin group compared to the placebo group after 4 weeks of treatment. These criteria improved by 12–38% in comparison to baseline in the verum group. Only slight improvements of up to 7% were found in the control group, which were not statistically significant.

Table 1. Effects of 4 weeks treatment (5 days per week) of DAT patients with 30 ml Cerebrolysin or placebo, and the long term effects after 6 months with respect to the clinical symptomatology (SCAG), cognitive performance in the trail-making test (ZVT-G), the activities of daily living (NAI) and the self assessment according to von Zerssen and Möller (BF-S). Two subgroups are also reported on the basis of the presence or absence of additional treatment received during the follow-up period. Average ± SD.; *$p < 0.05$; n = 49 (Cerebrolysin) / 52 (placebo)

		Control n = 60	4 weeks n = 60	Follow-up 28 weeks n = 49/52	No additional therapy n = 31/37	Additional therapy n = 18/15
ZVT-G	Cerebrolysin	184.1 ± 32.4	161.5 ± 22.8*	147.7 ± 17.0	146.3 ± 17.4	151.3 ± 16.1
	Placebo	184.4 ± 39.3	185.6 ± 21.5	162.0 ± 11.4	162.9 ± 14.8	159.4 ± 11.2
SCAG	Cerebrolysin	66.5 ± 6.5	49.8 ± 5.2*	54.2 ± 5.9*	56.8 ± 4.6	49.7 ± 5.3
	Placebo	66.9 ± 7.5	65.8 ± 6.1	63.7 ± 5.9	65.8 ± 4.3	58.3 ± 6.0
NAI	Cerebrolysin	48.1 ± 2.2	34.5 ± 2.1*	36.4 ± 3.4	37.8 ± 2.9	34.1 ± 2.8
	Placebo	48.2 ± 2.7	45.6 ± 3.3	44.0 ± 3.9	45.4 ± 2.1	40.5 ± 5.0
Bf-S	Cerebrolysin	43.7 ± 4.3	26.9 ± 6.7*	27.2 ± 3.6	26.9 ± 3.5	27.7 ± 3.9
	Placebo	44.7 ± 3.6	41.6 ± 5.7	27.4 ± 3.7	27.4 ± 3.3	27.5 ± 4.5

After 28 weeks the results indicated a further improvement of cognitive performance in the trail-making test from 8% to 20% in the Cerebrolysin group, compared to an overall improvement of 12% in the control group. There was no difference between the two sub-groups which received additional therapy during the follow-up period.

The improvements in clinical symptomatology (SCAG-score) are shown in Fig. 1. Whereas symptomatology improved by only 3% in the placebo group after treatment, and after the follow-up period by 5% cumulatively,

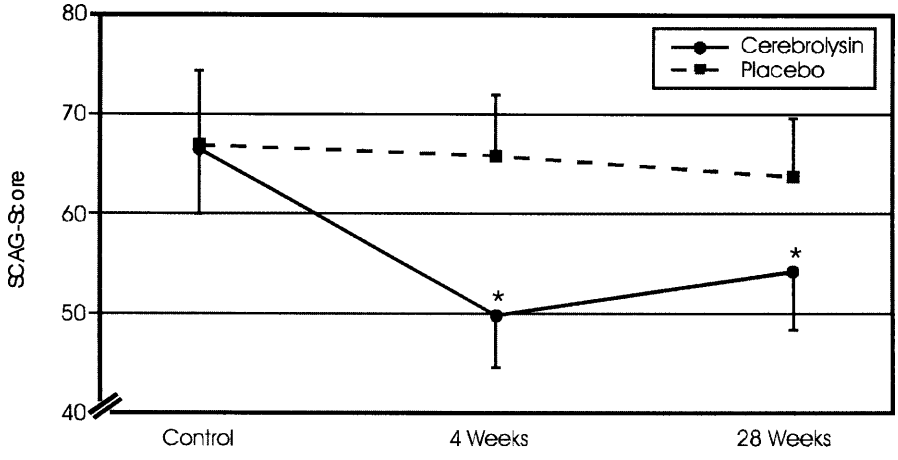

Fig. 1. Effects of 4 weeks treatment (5 days per week) of DAT patients with 30 ml Cerebrolysin or placebo, and the long term effects after 6 months with respect to the clinical symptomatology (SCAG-score). Average ± SD.; *$p < 0.05$; n = 49 (Cerebrolysin) / 52 (placebo)

Table 2. Effects of 4 week treatment (5 days per week) of DAT patients with 30 ml Cerebrolysin or placebo, and the long term effects after 6 months. Negative values at 4 week and follow up indicate an improvement in clinical symptomatology (SCAG-score)

SCAG-score	Cerebrolysin						Placebo					
Cluster	1	2	3	**4**	5	6	1	2	3	**4**	5	6
n	4	3	3	**30**	4	5	1	1	2	**35**	6	4
Baseline mean	55.8	60.3	64.0	**66.1**	76.8	74.8	7.6	56.0	62.5	**64.5**	73.0	76.5
Week 4 mean	−9.3	−5.0	−7.7	**−17.4**	−24.5	−24.4	−7.0	−6.0	0.0	**−0.4**	−0.3	−5.0
Follow-up mean	8.5	−8.3	−0.7	**6.3**	−6.8	9.0	6.0	9.0	−9.5	**1.3**	−15.7	−3.8

superior results were obtained in the Cerebrolysin group with 25% improvement after treatment and still 19% after 6 months. A slight trend was recognizable in the SCAG-score results indicating a small profit due to additional therapy during the follow-up period. Also in this case the results were in favor of Cerebrolysin. In general these results are in accordance with the expectation of non-specific changes in the placebo group and a slight decline in the verum group, compared to results immediately after end of the active treatment course. Changes between baseline and the follow-up examination, within the drug group and in comparison to the placebo group, display further significant ($p < 0.05$) improvements due to Cerebrolysin therapy.

Patients in the Cerebrolysin group who were more severe at baseline show a clear trend to a better outcome in clinical symptomatology after 28 weeks. On the other hand patients who had a good outcome at end of therapy (week 4) declined slightly in SCAG-score during the follow up period, still showing a net treatment profit compared to baseline. In the placebo group there was no such correlation. A low SCAG-score at baseline indicates milder severity than higher scores. Placebo patients in cluster 3, 4 and 5 show nearly no response after week 4 and the remaining three clusters worsened slightly (Table 2). The Cerebrolysin patients in cluster 4, 5 and 6 show a pronounced response after 4 weeks. Patients in the first three clusters are weak responders. Most of the patients in the Cerebrolysin and placebo group were represented in cluster 4, and both showed deterioration during the follow up period. In relation to baseline the placebo group is deteriorated (+0.9 points at month 6). The improvement of the Cerebrolysin patients (−17.4 points) achieved after 4 weeks of active treatment was slightly reduced to a value of −11.1 after 28 weeks. In the Cerebrolysin group there was no cluster with a deterioration compared to baseline and the week 4 evaluation.

In the activities of daily living compared to baseline there was an improvement from 5% after 4 weeks to 9% after 28 weeks in the placebo group. In the Cerebrolysin group there was a slight regression of the NAI score after 28 weeks in relation to the 4 week result, but the final value of 24% was still

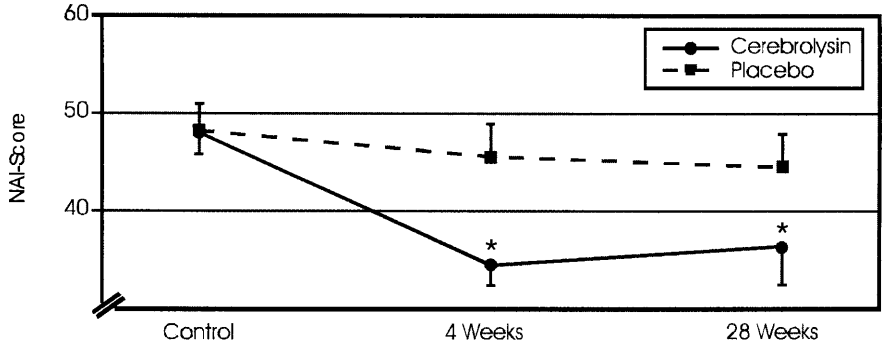

Fig. 2. Effects of 4 weeks treatment (5 days per week) of DAT patients with 30 ml Cerebrolysin or placebo, and the long term effects after 6 months with respect to the activities of daily living (NAI). Average ± SD.; *$p < 0.05$; n = 49 (Cerebrolysin) / 52 (placebo)

clearly above the value for the control group (Fig. 2) and the baseline. A trend favoring the sub-group which received additional therapy during the follow-up period has been observed. The effects were, however, more pronounced in the Cerebrolysin group than the placebo group.

The results of the von Zersson self-assessment test (Bf-S) of the Cerebrolysin group remained unchanged after 6 months, still with an improvement of 38% compared to baseline. The placebo group showed similar results after 28 weeks. No differences were observed between the sub-groups with and without additional treatment.

The Cerebrolysin group was superior to the control group with respect to Clinical Global Impressions upon completion of treatment as well as after the follow-up period ($p < 0.05$) (Table 3). Of the 49 patients (100%) in the Cerebrolysin group showing a positive change in condition after 4 weeks of medication, 46 (93.9%) maintained their improvements after 28 weeks. In contrast only 9 patients (17.3%) in the placebo group showed an improvement after the end of treatment, and 8 patients (15.4%) after the follow-up period. In the treatment group there was a decline from "much improved" to "minimally improved," however the general improvement was maintained.

With the exception of 1 case of bronchial pneumonia lasting 12 days (placebo group) there were no incidents of serious illness or other adverse events during the follow-up period. During the 4 week therapy there were no changes in blood-pressure or heart rate, and the laboratory parameters were not influenced in any clinically relevant manner. In addition to the clinical efficacy, Cerebrolysin was distinguished by its excellent tolerability, a fact already shown also in a series of other studies (Koppi and Barolin, 1996; Biesenbach et al., 1997; Rainer et al., 1997).

Discussion

At the present time there is no effective pharmacological therapy available which can either cure Alzheimer's disease or stop the progression. The

Table 3. Effects of 4 weeks treatment (5 days per week) of DAT patients with 30 ml Cerebrolysin or placebo, and the long term effects after 6 months with respect to the Clinical Global Impressions. Average ± SD.; *$p < 0.05$; n = 49 (Cerebrolysin) / 52 (placebo)

Change in condition (CGI) after 4 weeks

	Cerebrolysin*		Placebo	
	%	n	%	n
much improved	61.2	30	0.0	0
minimally improved	38.8	19	17.3	9
unchanged	0.0	0	82.7	43

Change in condition (CGI) after 28 weeks

	Cerebrolysin*		Placebo	
	%	n	%	n
much improved	30.6	15	3.9	2
minimally improved	63.3	31	11.5	6
unchanged	6.1	3	69.2	36
somewhat worse	0.0	0	15.4	8

dramatic increase in the prevalence of this illness in the next decades will lead to an enormous strain on the world's social systems. The duration of affliction is up to 20 years, whereby the first half of this period is characterized by milder symptoms. Gradually the patients lose their functional independence, and need intensive health care, which is often complicated by the development of accompanying behavioral symptoms (Reisberg et al., 1991).

In spite of enormous research efforts to develop efficacious pharmacological treatment for Alzheimer's disease until now only symptomatic drugs are available. Modern cholinesterase inhibitors like Donepezil (Doody, 1999) or Rivastigmine (Anand et al., 1996) are well tolerated and safe, providing reproducible improvements of cognitive performance and global funtioning of DAT patients. From the long-term experience it can be concluded that their use delays the progression of the disease for 6 to 12 months, but they do not provide a real disease modifying activity as shown by drug withdrawal experiments (Rogers et al., 1998). There are clinical studies available, reporting about disease modifying activity of high dose vitamin E treatment (Sano et al., 1996, 1997) using novel clinical outcome variables, showing a delay in nursing home placement or loss of activities of daily living, but failing to achieve cognitive effects. Also studies with the drug Propentofylline (Marcusson et al., 1997; Rother et al., 1998) indicate a disease modifying activity via influence on microglia activation, showing at least modest but significant improvements of cognitive and performance global function.

Preventive strategies such as the use of anti-inflammatory drugs (Breitner, 1996) or estrogens (Kawas et al., 1997; McBee et al., 1997) provide promising perspectives, however prospective, large scale clinical studies have to be completed. It was expected that the use of different neurotrophic factors, with specific focus on NGF, would open an efficient possibility to influence the progression of Alzheimer's disease (Gauthier et al., 1991; Lapchak, 1993; Hefti, 1999). First treatment trials in human beings resulted in inconclusive effects on cognition (Olson, 193; Seiger et al., 1993) and further studies were not performed until now, because of intolerable side effects, like the induction of hyperalgesia. Considering the recent information on drug effects in Alzheimer's disease the sustained improvements achieved with Cerebrolysin in the present study are of interest, especially because they were achieved after the relatively short treatment period of only 4 weeks or just 20 drug applications respectively (Rüther et al., 1994). In the original study the placebo group did not change at all over the whole 4-week treatment period, which was unexpected, because initial placebo response is a well known phenomenon in this kind of studies. On the other hand there was a quite uniform response in the Cerebrolysin group in all investigational parameters. The fact that these improvements could be maintained over a period of 6 months without treatment is surprising and can be explained only by the proposed neurotrophic activity of Cerebrolysin (Shimazu et al., 1992; Satou et al., 1994). However a decline in all different evaluation parameters was observed, compared to the results immediately after active treatment. This indicates that the drug is able to induce long lasting effects, but as expected, a single treatment course cannot stop the disease progression. In comparison the changes in the placebo group during the whole 7 months of observation were modest, and some improvements during the follow up could be attributed to two single patients displaying pronounced improvements. The only explanation for this phenomenon could be that both of the patients did not really suffer from Alzheimer's disease. However there was a clear distinction between the drug treated and the placebo groups at the end of the follow up, and the patients who received Cerebrolysin initially performed still significantly better compared to baseline. Unfortunately a small proportion of the patients received additional treatment with different nootropic drugs during the follow up period, but this did obviously not influence the overall group outcome at the final evaluation, with exception of small trends concerning improvements in clinical symptomatology and activities of daily living. The persisting improvements are in accordance with the described neurotrophic effect, and it is supported by reports about protection of cholinergic neurons after fimbria fornix transection in rats, a well described model for chronic neurodegeneration (Akai et al., 1992). Treatment after this lesion had an immediate influence on spatial learning and memory documented in experiments using the Morris Water Maze (Francis et al., 1996), but also displayed positive long-term effects on cognitive performance (Valouskova, 1998). Behavioral improvements were also reported in APOE-knock out mice (Masliah et al., 1999). In this animals a further consequence of Cerebrolysin application for 4 weeks was a complete normalization of the

dendritic cytoarchitecture which corresponded with elevated MAP-2 levels, and an enhanced synaptophysin immunoreactivity indicated an increased number of synapses. Also in 24-month-old rats in which Cerebrolysin improved memory and learning (Gschanes et al., 1998) clear morphological changes were described by Reinprecht et al. (1999), showing an increased synaptic density in the hippocampus and the entorhinal cortex. All these phenomena could explain the long lasting effects found in this clinical trial. Maybe an additional benefit can be achieved by the ability of Cerebrolysin to increase functional blood-brain barrier glucose transporter GLUT-1 (Boado et al., 1999). It is known that this rate limiting transporter protein is reduced in Alzheimer's disease (Mooradian et al., 1997), which might cause an additional neuronal energy deficit, culminating in metabolic disturbances contributing to the overall degenerative process. Also the recently described effect preventing microglia activation and production of cytokines, like Interleukin-1β, is in accordance with the clinical data indicating a pronounced influence on disease progression.

However the relatively short overall observation period of only seven months does not allow definite conclusions about a disease modifying effect of Cerebrolysin. It is interesting to see that a neurotrophic treatment approach, which was obviously provided with Cerebrolysin, had quite pronounced effects especially on activities of daily living, being most relevant for the quality of life of the patients and their care givers. For final conclusions about therapeutic relevance of this drug in Alzheimer's disease a prolongation of the observation period to at minimum one year, preferentially even longer, is necessary. Also consequences of repeated treatment courses would be of interest.

It can be summarized that Cerebrolysin seems to provide a disease modifying treatment possibility based on neurotrophic activity. The promising data of the present study have to be explored in further long-term trials.

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