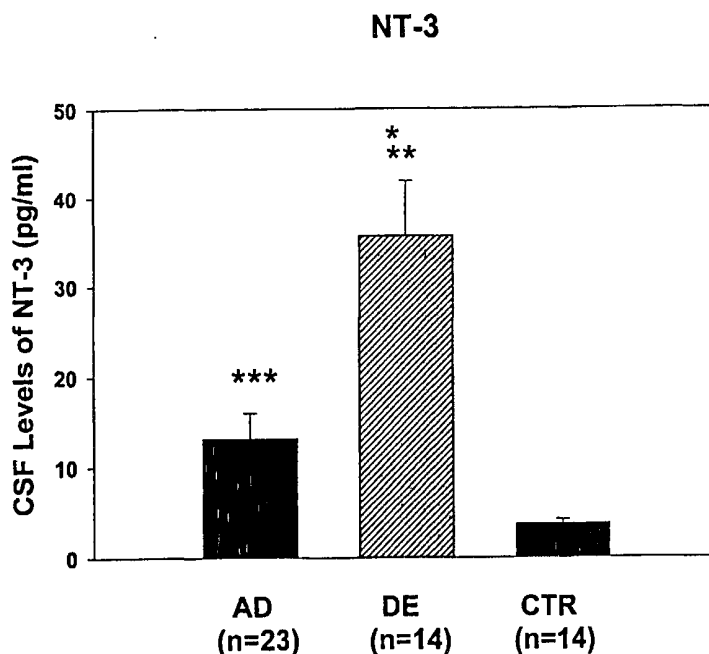




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(54) Title: METHODS OF DIAGNOSING OR TREATING DEPRESSION



(57) Abstract

The invention relates to a method for diagnosing or prognosing depression, in particular major depression, in a subject, or determining whether a subject is at increased risk of developing depression, in particular major depression, comprising: determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject; and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status, thereby diagnosing or prognosing depression in said subject, or determining whether said subject is at increased risk of developing said depression.

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METHODS OF DIAGNOSING OR TREATING DEPRESSION

The present invention relates to methods of diagnosing or treating depression as well as to screening methods for identifying suitable agents to be used in the treatment of depression.

In the search for biochemical changes in patients with neuropsychiatric or neurodegenerative disorders analysis of cerebrospinal fluid (CSF) may be a useful method, since the CSF is continuous with the extracellular fluid of the brain. Therefore, a plurality of studies aiming at the analysis of the central nervous system (CNS) specific proteins in CSF were performed in order to find biochemical markers for neuronal and synaptic function and pathology in degenerative brain disorders.

Neurotrophin 3 (NT-3) is a member of the neurotrophin gene family which supports the survival, differentiation, maintenance, and repair of vertebrate neurons (Bothwell, *Ann. Rev. Neurosci.* 8, 223 - 253, 1995; Ebadi et al., *Neurochemistry International* 30 (4-5), 347-374, 1997). It was shown to prevent the death of adult central noradrenergic neurons of the locus coeruleus in a 6-hydroxydopamine lesion model (Arenas et. al., *Nature* 367 (6461), 368 - 371, 1994), a neuronal population which is associated with the pathophysiology of major depression (Leonard, *J. Psychopharmacol.*, 11 (4), 39 - 47, 1997). Alterations in adrenoceptor density and function, and changes in adrenoceptors associated with the pituitary-adrenal axis function strongly implicate a disorder in central noradrenergic transmission in major depression (for review see: Leonard, *J. Psychopharma* 11 (4), 39 - 47, 1997). This dysfunction might be related to the activity of tyrosine hydroxylase, the rate-limiting enzyme in the synthesis of catecholamines. Alternatively,

impaired uptake or transport of a target-derived neurotrophic factor, such as NT-3, may contribute to central noradrenergic dysfunction.

The international patent application PCT/US 90/04916 relates to NT-3 and its use in the diagnosis and/or treatment of neurologic disorders including peripheral neuropathies such as diabetic neuropathies, toxic and nutritional neuropathies, hereditary neuropathies and AIDS related neuropathies and degenerative diseases as Alzheimer's disease.

Murase et al. (*Clinica Chimica Acta*, 227(1-2), 23 - 36, 1994) developed an enzyme immunoassay system for NT-3, based on a biotin-streptavidin detection system. This system was applied to measure NT-3 levels in the developing rat nervous system as well as in normal subjects and patients with Alzheimer's disease.

Using two-site enzyme immunoassays, Narisawa-Saito et al. (*Neuroreport* 7(18), 2925 - 2928, 1996) measured inter alia NT-3 in three brain regions (motor cortex, dentate gyrus and entorhinal cortex) of patients with Alzheimer's disease and control individuals.

The international patent application PCT/US 92/09652 provides for therapeutic and diagnostic method based on human NT-3 expression, specifically the potential to treat Alzheimer's disease and Huntington's disease.

NT-3 was previously determined in the CSF of patients with a variety of neuropsychiatric conditions including hydrocephalus, meningitis, encephalitis, ventriculitis, brain tumors, and multiple sclerosis, with values ranging from 4.8 to 55.9 pg/ml (Gilmore et al., *Psychiatry Res.* 73(1-2) 109 - 113, 1997). In the report of Gilmore, NT-3 was not detectable in the CSF of patients with schizophrenia.

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Up to date, studies on CSF levels of NT-3 in major depression in the elderly (DE) or in depressive populations of younger age have not yet been reported.

Despite its relevance for quality care initiatives, the field of psychiatry has little scientific knowledge regarding the course of current major depression when primary care patients with the disorder remain undetected. The risk that undetected patients develop suicidal ideation should not be underestimated. Therefore, methods of diagnosing depression, in particular major depression, as well as methods of treatment are urgently needed.

In one aspect, the invention features a method for diagnosing or prognosing depression, in particular major depression, in a subject, or determining whether a subject is at increased risk of developing depression, in particular major depression, comprising:

- determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject;

- and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status,

- thereby diagnosing or prognosing depression in said subject, or determining whether said subject is at increased risk of developing depression.

In a further aspect, the invention features a method of monitoring the progression of depression, in particular major depression, in a subject, comprising:

- determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject;

- and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status,

- thereby monitoring the progression of depression in said subject.

In still a further aspect, the invention features a method of evaluating a treatment for depression, in particular major depression, comprising:

determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject;

and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status, thereby evaluating said treatment for depression.

It is preferred to use a body fluid, preferably cerebrospinal fluid, as said sample. An increase of a level of neurotrophin 3 in said cerebrospinal fluid from said subject relative to said reference value representing a known health status indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject. In particular, a level of neurotrophin 3 ≥ 15 pg/ml in CSF indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject. Specifically, a level of neurotrophin 3 in the range from 15 pg/ml to 50 pg/ml in CSF indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject. The increases in CSF levels of NT-3 may reflect disturbances in the trophic support of specific neuronal populations, such as the central noradrenergic system in DE.

It is particularly preferred to compare a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in said sample with a level of at least one of said substances in a series of samples taken from said subject over a period of time. Preferably said subject, e.g. a mammalian such as a human, receives a treatment prior to one or more of said sample gatherings. Neurotrophin 3 and/or said transcription product can preferably be detected using an immunoassay and/or a binding assay.

In another aspect, the invention features a kit for diagnosis, or prognosis, or determination of increased risk of developing depression, in particular major depression, in a subject, said kit comprising:

- (a) at least one reagent which selectively detects neurotrophin 3 or a transcription product of a gene coding for neurotrophin 3; and
- (b) instructions for diagnosing, or prognosing depression, or determining increased risk of developing depression by
 - (i) detecting a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject; and
 - (ii) diagnosing, or prognosing, or determining whether said subject is at increased risk of developing depression,

wherein an varied level, or activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 compared to a reference value representing a known health status;

or a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 similar or equal to a reference value representing a known disease status

indicates a diagnosis, or prognosis, or increased risk of developing depression.

In particular, a level of neurotrophin 3 ≥ 15 pg/ml in CSF indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject. Specifically, a level of neurotrophin 3 in the range from 15 pg/ml to 50 pg/ml in CSF indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject. Additionally, said kit preferably further comprises at least one reagent which selectively detects nerve growth factor or a transcription product of a gene coding for nerve growth factor. Combined testing of NT-3 and NGF is a valuable tool in the diagnosis, prognosis, or risk evaluation of the above mentioned diseases (see example 3 and table 1).

In still another aspect, the invention features a method of treating or preventing depression, in particular major depression, in a subject comprising administering to said subject in a therapeutically effective amount an agent or agents which directly or indirectly affect, in particular reduce or increase, an activity, or a level, or both said activity and level, of at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3.

In still another aspect, the invention features a use of an agent for the manufacture of a medicament for treating depression, in particular major depression, wherein said agent directly or indirectly affects, in particular reduces or increases, an activity, or a level, or both said activity and level, of at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3.

In still another aspect, the invention features a method for identifying an agent that directly or indirectly affects an activity, or a level, or both said activity and level, of at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3, comprising the steps of:

- (a) providing a sample containing at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3;
- (b) contacting said sample with at least one agent;
- (c) comparing an activity, or a level, or both said activity and level, of at least one of said substances before and after said contacting.

By means of this assay principle, agents can be identified which are suitable drugs or precursors of such drugs in a therapy of depression, in particular major depression.

Figure 1 relates to example 1 and depicts that CSF levels of NT-3 were significantly elevated in the DE group, as compared to both the AD and the CTR groups. Levels (pg/ml) are given in mean \pm SEM. Asterisk (*, **, ***) indicate significance ($p < 0.005$), Mann-Whitney U Test. NT-3: * DE versus AD, $p = 0.004$; ** DE versus CTR, $p < 0.001$; *** AD versus CTR, $p = 0.013$. NT-3 concentrations in CSF of the DE group were 35.7 ± 6.2 pg/ml (mean \pm SEM; range 2.3 to 87.00, $n = 14$), compared to 13.2 ± 2.8 pg/ml in the AD group (range: 2.0 to 41.0, $n = 23$), and 3.7 ± 0.5 pg/ml in the CTR group (range: 0.00 to 8.2, $n = 14$), respectively. In addition, CSF levels of NT-3 were significantly higher in the AD group as compared to the CTR group. There was no apparent correlation of CSF levels of NT-3 with age, duration of AD, MMS, NOSGER or MADRS scores.

Figure 2 relates to example 1 and depicts CSF levels of NT-3 (pg/ml \pm SEM) grouped for presence of ApoE4 alleles. Left panel: Alzheimer's disease group, $n = 22$, Kruskal-Wallis: $\chi^2 = 7.495$, $p = 0.024$. Right panel: total sample of patients with AD and major depression (DE), $n = 36$, Kruskal-Wallis test: $\chi^2 = 6.589$, $p = 0.037$. CSF levels of NT-3 increased with increasing ApoE4 allele frequency in the total sample of AD and DE patients and within the AD group. There was no apparent effect of the number of E4 alleles within the DE group ($n = 14$, Kruskal-Wallis test: $\chi^2 = 1.508$, $p = 0.471$).

Figure 3 relates to example 2 and depicts NT-3 levels in the cerebrospinal fluid of patients with Alzheimer's disease (AD), major depression in the elderly (DE) and non-demented control subjects (CTR). Levels (pg/ml) are given in mean \pm SEM. Asterix (*, **, ***) indicate significance ($p < 0.05$), Mann-Whitney U Test. * DE versus AD, $p = 0.005$; ** DE versus CTR, $p = 0.000$; *** AD versus CTR, $p = 0.010$. CSF levels of NT-3 were significantly elevated in the

DE group, as compared to both the AD and the CTR group. CSF levels of NT-3 were slightly, but significantly, elevated in the AD group, as compared to the CTR group. NT-3 concentrations in CSF of the DE group were 25.8 +/- 4.3 pg/ml (mean +/- SEM, range: 0.0 to 87.0, n = 23), compared to 14.0 +/- 1.6 pg/ml in the AD group (range: 0.0 to 41.0, n = 39), and 10.5 +/- 1.6 pg/ml in the CTR group (range: 0.0 to 67.0, n = 63), respectively.

Figure 4 relates to example 3 and depicts nerve growth factor (NGF) levels in the cerebrospinal fluid of patients with Alzheimer's disease (AD), major depression in the elderly (DE) and non-demented control subjects. CSF levels of NGF were determined to define a cut-off value to be used in the combined tests shown in table 1. Levels (pg/ml) are given in mean \pm SEM. Asterix (*, **, ***) indicate significance ($p < 0.05$), Mann-Whitney U test. * AD versus DE, $p = 0.002$; ** AD versus CTR, $p = 0.000$, *** DE versus CTR, $p = 0.000$. CSF levels of NGF were significantly elevated in the AD group, as compared to both the DE and the CTR group. CSF levels of NGF were also significantly elevated in the DE group, as compared to the CTR group. NGF concentrations in CSF of the AD group amounted to 8.19 +/- 0.91 pg/ml (mean +/- SEM, range: 0.00 to 23.00, n = 40), compared to 4.26 +/- 0.97 pg/ml in the DE group (range: 0.00 to 23.00, n = 22), and 1.18 +/- 0.35 pg/ml in the CTR group (range: 0.00 to 7.20, n = 32), respectively.

Figure 5 depicts CSF levels of NT-3 in DE patients (n = 23) grouped according to treatment with antidepressants: substances that affect central noradrenergic neurotransmission (MAOI and TCA, n = 14); substances that selectively affect serotonergic neurotransmission (SSRI, n = 6); no treatment with antidepressants at the time of lumbar puncture (N=3). CSF levels of NT-3 (pg/ml) are given in mean \pm SEM. Asterix (*) indicates significance ($p < 0.05$, independent samples *t* - test, MAOI/TCA versus SSRI). Interestingly, Smith et al. (Proc. Natl. Acad. Sci. USA, 92 (19), 8788 - 8792, 1995) showed that chronic treatment with antidepressants that selectively blocked neuronal noradrenaline uptake decreased the expression levels of NT-3 in the locus

coeruleus in rats. In contrast, treatment with selective serotonin reuptake inhibitors did not alter NT-3 mRNA levels. These findings suggest that some effects of antidepressants on the function of the locus coeruleus involve NT-3 expression. However, these findings make it highly unlikely that the increased CSF levels of NT-3 in patients with DE are due to effects of treatment with antidepressants. In fact, CSF levels of NT-3 were markedly lower within the DE group in patients treated with substances that affect central noradrenergic neurotransmission (MAOI/TCA) than in patients treated with substances that selectively affect serotonergic neurotransmission (SSRI) as well as in patients that were not treated with antidepressants at the time of lumbar puncture. Thus, treatment with antidepressants that block neuronal noradrenaline uptake may reduce previously increased CSF levels of NT-3 in patients with DE.

Table 1 relates to examples 2 and 3. This table shows the diagnostic accuracy of spinal fluid measurements of NT-3 and NGF in Alzheimer's Disease and Major Depression in the elderly. In NT-3 measurements, sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) were calculated using a cut-off value of ≥ 15.0 pg/ml (total: n = 125, AD: n = 39, DE: n = 23, CTR: n = 63). For combined NT-3/NGF measurements in the diagnosis of DE, cut-off values of < 4 pg/ml NGF, and ≥ 15 pg/ml NT-3, respectively (total: n = 57, AD: n = 24, DE: n = 18, CTR: n = 15) were used. CSF measurement of NT-3 levels showed a promising diagnostic accuracy in terms of sensitivity (73.9 %) as well as specificity (86.1 %). Thus, the NT-3 test constitutes a candidate tool for the biochemical diagnosis of major depression in the elderly. In the combined test for DE diagnosis, the specificity is slightly increased (89.7 %). For combined NT-3/NGF measurements in the diagnosis of AD, cut-off values of < 15 pg/ml NT-3 and ≥ 4 % pg/ml NGF, respectively (total n = 57; AD n = 24, DE n = 18, CTR n = 15) were used. This combined test shows a considerable specificity for the diagnosis of AD (90.1 %). In general, combined testing of NT-3 and NGF with suitable cut-off values

has the potential to be used in the biochemical differentiation between these two frequent disorders in the elderly.

EXAMPLE 1

In order to achieve a differential diagnosis, the study included not only patients with major depression (DE), but also such with Alzheimer's disease. Patients with major depression were diagnosed according to the ICD10 (F32.0x/1x, F33.0x/1x) and DSM-III-R (296.20-22, 296.30-32) criteria. Diagnosis of probable AD was made according to criteria of the National Institute of Neuropsychiatric and Communicative Disorders and Stroke-Alzheimer's disease and Related Disorders Association (NINCDS-ADRDA; McKhann et al., *Neurology* 34, 939 – 944, 1984). All patients were referred to the research ward from general practitioners, neurologists and psychiatrists for diagnostic purposes and screening for clinical trials. None of the patients was institutionalized. The group of healthy control subjects (CTR) consisted of patients that underwent lumbar puncture for orthopedic or neurologic diagnostic purposes and were shown to have normal CSF cell counts, total protein levels, and absence of signs of blood barrier dysfunction or cerebral IgG synthesis, as well as absence of any cerebral disorders.

DE, AD and CTR patients were carefully examined and received a thorough clinical work-up. Psychometric testing including the Mini Mental State (MMS; Folstein et al., *J. Psychiatry Res.* 12, 189 – 198, 1975), as a global screening instrument for dementia, and the Nurses' Observation Scale for Geriatric Patients (NOSGER; Spiegel et al., *J. Am. Geriatr. Soc.* 39(4), 339 – 347, 1991) as a functional measure of dementia severity. The patients with DE showed no cognitive disturbances in the clinical examinations and the Mini Mental State scores were within the normal range. Severity of depression was rated by using the Montgomery Asberg Depression Rating Scale (MADRS) (Montgomery et al., *Br. J. Psychiatry*, 134: 382 – 389, 1979). Apolipoprotein (ApoE)

genotyping, or, if DNA was not available, ApoE phenotyping was included in the laboratory screening in the DE and AD patients.

CSF was obtained for diagnostic purposes in the DE and AD patients in which no lumbar puncture had been previously done during the routine diagnostic work-up. Different CSF volumes were available for the analysis of the neurotrophin proteins. This fact explains the different sample sizes for the individual measurements. All available CSF samples were used for the analyses.

The AD group was as follows: $n = 23$, 12 men, 11 women, mean age 63.9 ± 13.2 SD years, range 39 – 86 years, MMS score: mean 18.6 ± 5.6 SD.

The DE group was as follows: $n = 14$, 5 men, 9 women, mean age 68.2 ± 13.6 SD years, range 47 – 86 years, MMS score: mean 28.1 ± 0.9 SD.

The CTR group was as follows: $n = 14$, 8 men, 6 women, mean age 58.5 ± 16.2 SD years, range 31 – 81 years.

AD and CTR patients were free of psychotropic medication. Patients with major depression were treated with various antidepressant drugs including serotonin reuptake inhibitors, reversible monoaminooxidase A inhibitors and tricyclics. Informed consent was taken from each patient and their caregivers before the investigation. The study was approved by the local ethics committee. All procedures were in accordance with the Helsinki Declaration of 1975, as revised in 1983. Within one week of dementia testing, CSF was obtained by lumbar puncture. To control for possible influences of a ventriculo-lumbar gradient, lumbar punctures were done between 7.30 and 8 a. m. before breakfast while patients were still lying flat. CSF samples were frozen on dry ice immediately upon withdrawal at the bedside in 0.5 ml aliquots and stored at $-85\text{ }^{\circ}\text{C}$ until biochemical analysis.

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NT-3 was determined using commercially available ELISA systems (Promega, Madison, WI). The determination was performed according to the manufacturer's protocol. 220 μ l of undiluted CSF in carbonate buffer (pH 9.7) were added to 96 well immunoplates (Nunc) at 4 °C overnight. Anti-Human-NT-3 polyclonal antibodies (pAb) were used as capture antibody. Anti-NT-3 monoclonal antibody (mAb) were used as reporter antibody. After incubation with a species-specific Ab (anti-rat IgG) conjugated to horseradish peroxidase (HRP) as a tertiary reactant, and washing, the solution was incubated with the chromogenic substrate TMB (3, 5, 3', 5'-tetramethylbenzidine). Absorbance was measured at 450 nm using a microplate reader (Dynatech MR 700). NT-3 ELISA: range 4.7 – 300 pg/ml; cross-reaction with other neurotrophins at 10 ng/ml < 3 %; detection limit 10.0 pg/ml. All CSF samples were assayed in duplicate determinations.

ApoE genotyping was performed using INNO-LiPA ApoE, Innogenetics, Belgium. ApoE phenotyping was performed according to McDowell et al. (Clin. Chem. 35(10), 2070 – 2073, 1989). The use of the ApoE phenotype synonymous with the ApoE genotype in the statistical analyses seemed to be appropriate, since ApoE genotyping compared with protein phenotyping showed conflicting results in less than 2 % (Hansen et al., Clin. Chim. Acta, 224(2), 131 – 137, 1994).

Statistical analyses of data were performed using the Mann-Whitney U test as well as independent samples *t*-tests for group comparisons. Correlation analyses were performed by multiple regression using CSF levels of neurotrophins as well as ApoE genotype (or phenotype, respectively), age, duration of the disease in AD, MMS, NOSGER and MADRS scores. Non-parametric correlation was performed using the Kruskal-Wallis test. Regression analysis was complemented with analysis of variance (ANOVA) by using SPSS for Windows (version 8.0). Statistical significance was assumed at $p < 0.05$. Bonferroni correction for multiple testing was applied.

EXAMPLE 2

The study described in example 1 has been extended to a wider panel of patients as described below in this example 2.

Diagnosis, clinical examination and treatment of patients as well as lumbar puncture were performed as described in example 1.

For NT-3 measurements, 125 spinal fluid samples were examined. DE group: n = 23, 8 men, 15 women, mean age 70.5 +/- 11.9 SD years, range 47 - 86 yr, MMS score: mean 27.2 +/- 2.5 SD. AD group: n = 39, 20 men, 19 women, mean age 67.2 +/- 11.5 SD years, range 39 - 86 yr, MMS score: mean 19.1 +/- 5.3 SD. CTR group: n = 63, 35 men, 28 women, mean age 56.0 +/- 15.0 SD years, range 28 - 84 yr.

NT-3 was determined by using commercially available ELISA systems (Promega, Madison, WI) according to the manufacturer's protocol. 120 µl of undiluted CSF in carbonate buffer (pH 9.7) were added to 96 well immunoplates (Nunc) at 4 °C overnight. Anti-Human-NT-3 polyclonal antibodies (pAb) were used as capture Ab. Anti-NT-3 mAb were used as reporter Ab. After incubation with a species-specific Ab (anti-rat IgG) conjugated to horseradish peroxidase (HRP) as a tertiary reactant, and washing, the solution was incubated with the chromogenic substrate TMB. Absorbance was measured at 450 nm by using a microplate reader (Dynatech MR 700). NT-3 ELISA: linear range 4.7 - 300 pg/ml; cross-reaction with other neurotrophins at 10 ng/ml < 3%; detection limit 6.0 pg/ml.

AD and CTR patients were free of psychotropic medication. Patients with major depression were treated with various antidepressants: 11 were treated with tricyclics (TCA), 2 with a monoamine oxidase-A inhibitor (MAOI), 1 with a combination of TCA with MAOI, 6 with selective serotonin reuptake inhibitors (SSRI), and 3 were free of antidepressants at the time of lumbar puncture.

Statistical analyses of data were performed using the Mann-Whitney U test for group comparisons. Regression analysis was complemented with analysis of variance (ANOVA) by using SPSS for Windows (version 8.0). Statistical significance was assumed at $p < 0.05$. Bonferroni correction for multiple testing was applied.

To estimate the diagnostic accuracy of the test, a) sensitivities and b) specificities, defined as follows, were calculated: a) true positives / (true positives and false negatives), and b) true negatives / (true negatives and false positives). To estimate the probability of disease, predictive values of the tests were calculated. The positive predictive value (PPV) was defined as true positives / (true positives + false positives). The negative predictive value (NPV) was defined as true negatives / (true negatives + false negatives).

Next, DE patients ($n = 23$) were grouped according to treatment with substances that affect central noradrenergic neurotransmission (MAOI and TCA, $n = 14$) and those that selectively affect serotonergic neurotransmission (SSRI, $n = 6$). CSF levels of NT-3 were significantly lower in the MAOI/TCA group, as compared to the SSRI group ($p < 0.05$, independent samples t - test). NT-3 concentrations in CSF of the MAOI/TCA group were 19.7 ± 2.9 pg/ml (mean \pm SEM, range: 0.0 to 35.0, $n = 14$), compared to 34.9 ± 8.5 pg/ml in the SSR group (range: 14.0 to 63.0, $n = 6$), and 35.7 ± 26.3 pg/ml in the group with patients that were not treated with antidepressants at the time of lumbar puncture (range: 0.00 to 87.0, $n = 3$) (Fig. 5).

EXAMPLE 3

The purpose of this study was to check whether combined measurements of the CSF levels of NT-3 and nerve growth factor (NGF) – which also belongs to the group of neurotrophins – improves the diagnostic accuracy of the NT-3 test described in example 2.

In a first step, NGF levels in CSF were determined to define a cut-off value to be used in the combined tests. Diagnosis, clinical examination and treatment of patients, lumbar puncture and statistical analyses were performed as described in example 2. 94 spinal fluid samples were examined. The AD group (n= 40) consisted of 18 men and 22 women, mean age 68.8 +/- 12.4 years, range 39 – 88 yr, MMS score: mean 19.3 +/- 4.6 SD. The DE group (n = 22) consisted of 8 men and 12 women, mean age 69.8 +/- 12.6 SD years, range 47 – 86 yr, MMS score: mean 27.5 +/- 2.1 SD. CTR group: n = 32, 18 men, 14 women, mean age 64.0 +/- 14.9 SD years, range 29 – 96 yr. CSF levels of NGF were measured by an ELISA as described by Weskamp et al. (J. Neurochem. 48: 1779 – 1786, 1987). Black 96-well microplates (Nunc) were coated with monoclonal anti- β (2.5 S, 7S) NGF antibodies (Ab) (clone 27/21, Boehringer Mannheim) diluted in carbonate buffer pH 9.2 overnight at 4 °C. 120 μ l of CSF and standard solutions were added and incubated for 20 hours at 4 °C. Plates were washed and incubated with anti- β (2.5 S, 7S) NGF- β -galactosidase conjugate for 2 ½ hours at room temperature (RT). Following an additional washing step, the fluorogenic substrate 4-methylumbelliferyl- β -D-galactopyranoside was added and plates were incubated at 4 °C overnight. The reaction was stopped after 1h at RT, and the fluorescent product was measured in the microtiter wells by using a fluorometer (Labsystems Fluoroskan Ascent FL) at 355 nm excitation and 460 nm emission wavelength. The detection limit was 0.5 pg/ml; the cross-reactivity with other neurotrophins at 10 ng/ml was < 2 % and the assay was linear over a range of 0.5 to 500 pg/ml.

57 CSF samples were available for combined NGF/NT-3 measurements. AD group: n = 24, 13 men, 11 women, mean age 64.9 +/- 12.5 SD years, range 47 – 82 yr, MMS score: mean 18.6 +/- 5.4 SD. DE group: n = 18, 7 men, 11 women, mean age 69.5 +/- 12.7 SD years, range 47 – 84 yr, MMS score: mean 27.7 +/- 2.1 SD. CTR group: n = 15, 10 men, 5 women, mean age 59.0 +/- 15.9 SD years, range 29 – 80 yr.

CLAIMS

1. A method for diagnosing or prognosing depression, in particular major depression, in a subject, or determining whether a subject is at increased risk of developing depression, in particular major depression, comprising:
determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject;
and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status,
thereby diagnosing or prognosing depression in said subject, or determining whether said subject is at increased risk of developing depression.

2. A method of monitoring the progression of depression, in particular major depression, in a subject, comprising:
determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject;
and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status, thereby monitoring the progression of depression in said subject.

3. A method of evaluating a treatment for depression, in particular major depression, comprising:
determining a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject;

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and comparing said level, or said activity, or both said level and said activity, to a reference value representing a known disease or health status,

thereby evaluating said treatment for depression.

4. The method according to any of claims 1 to 3, wherein said sample is a body fluid, preferably cerebrospinal fluid.
5. The method according to claim 4, wherein an increase of said level of neurotrophin 3 in said cerebrospinal fluid from said subject relative to said reference value representing a known health status indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject.
6. The method according to claim 5, wherein a level of neurotrophin 3 \geq 15 pg/ml in said cerebrospinal fluid indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject.
7. The method according to claim 6, wherein a level of neurotrophin 3 in the range from 15 pg/ml to 50 pg/ml in said cerebrospinal fluid indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject.
8. The method according to any of claims 1 to 7, wherein said subject is a human.
9. The method according to any of claims 1 to 8, wherein neurotrophin 3 and/or said transcription product is detected using an immunoassay and/or a binding assay.
10. The method according to any of claims 1 to 9, further comprising comparing a level, or an activity, or both said level and said activity, of

neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in said sample with a level, or an activity, or both said level and said activity, of at least one of said substances in a series of samples taken from said subject over a period of time.

11. The method according to claim 10, wherein said subject receives a treatment prior to one or more of said sample gatherings.

12. The method according to claim 11, wherein said level, or said activity, or both said level and said activity, in said samples is determined, before and after said treatment of said subject.

13. A kit for diagnosis, or prognosis, or determination of increased risk of developing depression, in particular major depression, in a subject, said kit comprising:

- (a) at least one reagent which selectively detects neurotrophin 3 or a transcription product of a gene coding for neurotrophin 3; and
- (b) instructions for diagnosing, or prognosing depression, or determining increased risk of developing depression by
 - (i) detecting a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 in a sample from said subject; and
 - (ii) diagnosing, or prognosing, or determining whether said subject is at increased risk of developing depression,

wherein an varied level, or activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 compared to a reference value representing a known health status;

or a level, or an activity, or both said level and said activity, of neurotrophin 3 and/or of a transcription product of a gene coding for neurotrophin 3 similar or equal to a reference value representing a known disease status

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indicates a diagnosis, or prognosis, or increased risk of developing depression.

14. The kit according to claim 13, wherein an increase of a level of neurotrophin 3 in cerebrospinal fluid from said subject relative to a reference value representing a known health status indicates a diagnosis, or prognosis, or increased risk of depression.
15. The kit according to claim 14, wherein a level of neurotrophin 3 ≥ 15 pg/ml in cerebrospinal fluid indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject.
16. The kit according to claim 15, wherein a level of neurotrophin 3 in the range from 15 pg/ml to 50 pg/ml in cerebrospinal fluid indicates a diagnosis, or prognosis, or increased risk of depression, in particular major depression, in said subject.
17. The kit according to any of claims 13 to 16 for use in monitoring a progression of depression in a subject.
18. The kit according to any of claims 13 to 16 for use in monitoring success or failure of a therapeutic treatment of a subject.
19. A method of treating or preventing depression, in particular major depression, in a subject comprising administering to said subject in a therapeutically effective amount an agent or agents which directly or indirectly affect an activity, or level, or both said activity and level, of at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3.

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20. Use of an agent for the manufacture of a medicament for treating depression, in particular major depression, wherein said agent directly or indirectly affects an activity, or a level, or both said activity and level, of at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3.
21. A method for identifying an agent that directly or indirectly affects an activity, or a level, or both said activity and level, of at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3, comprising the steps of:
- (a) providing a sample containing at least one substance which is selected from the group consisting of a gene coding for neurotrophin 3, a transcription product of a gene coding for neurotrophin 3, and neurotrophin 3;
 - (b) contacting said sample with at least one agent;
 - (c) comparing an activity, or a level, or both said activity and level, of at least one of said substances before and after said contacting.

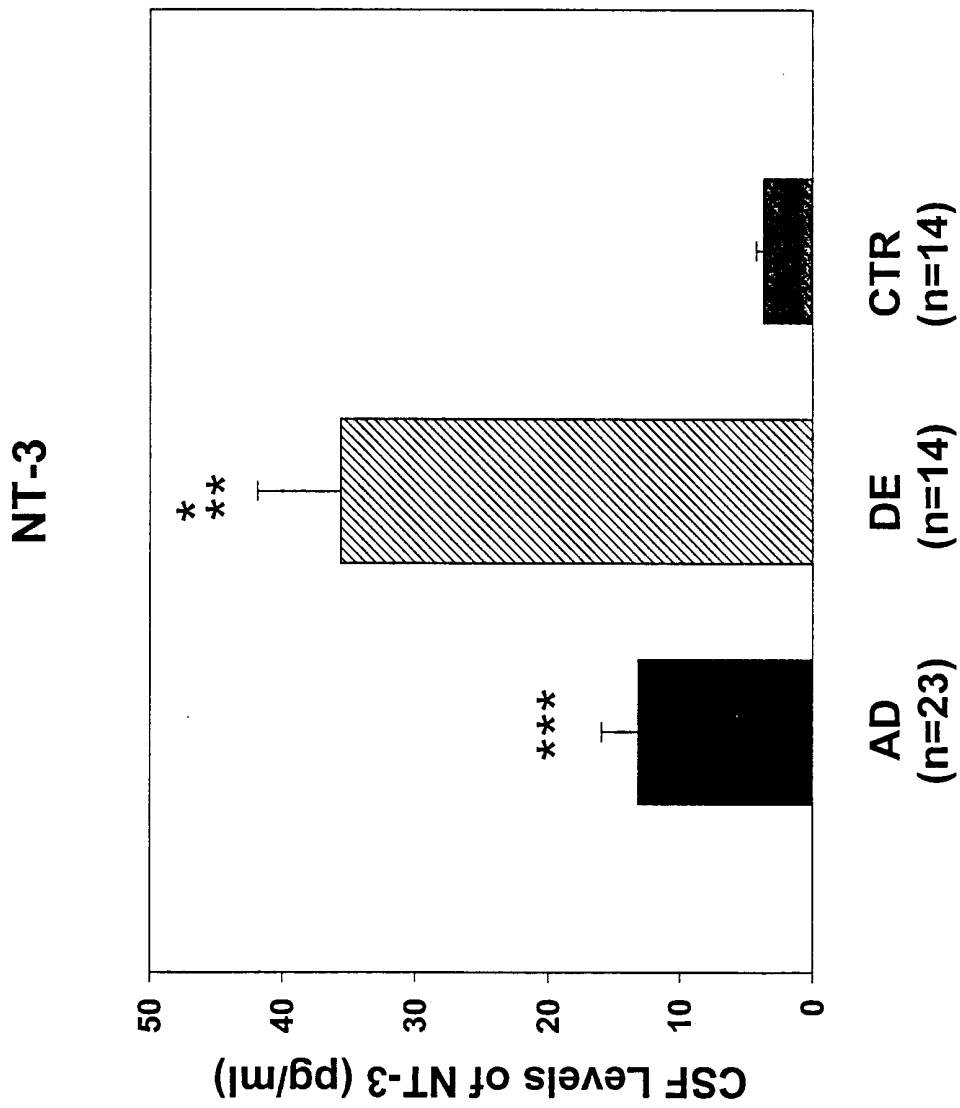


Fig. 1

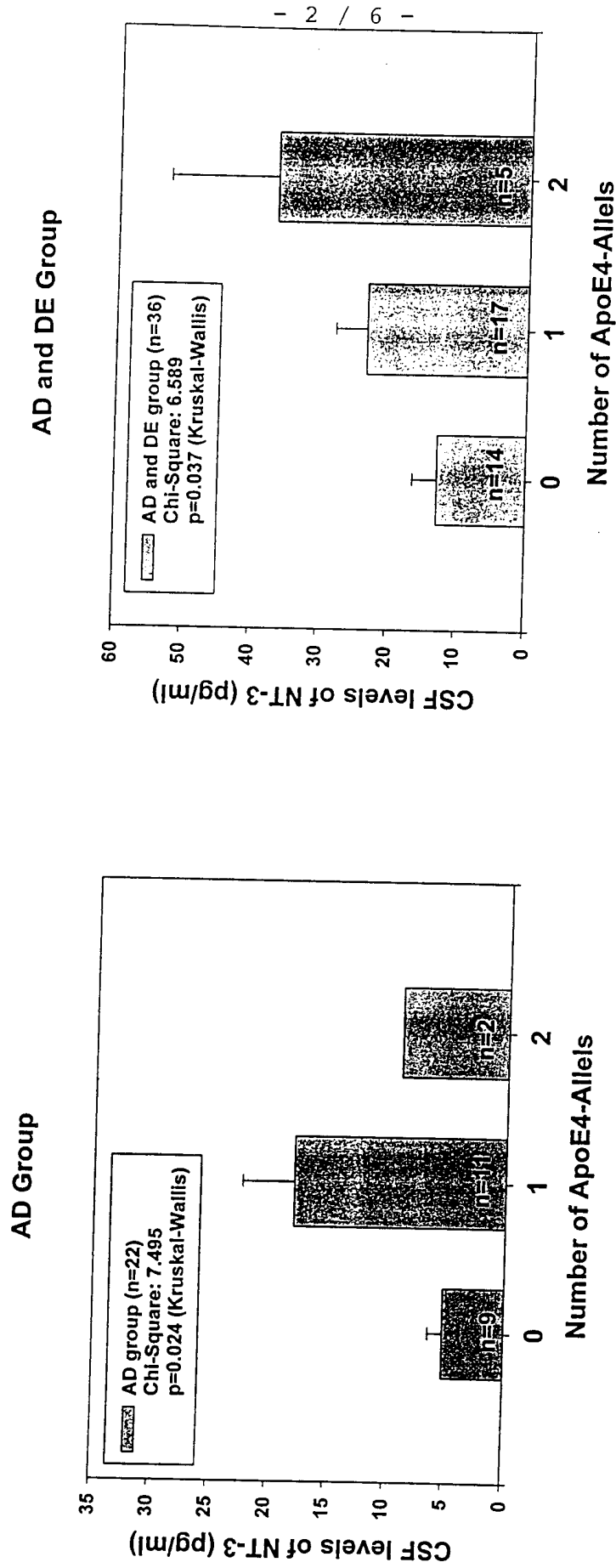


Fig. 2

NT-3

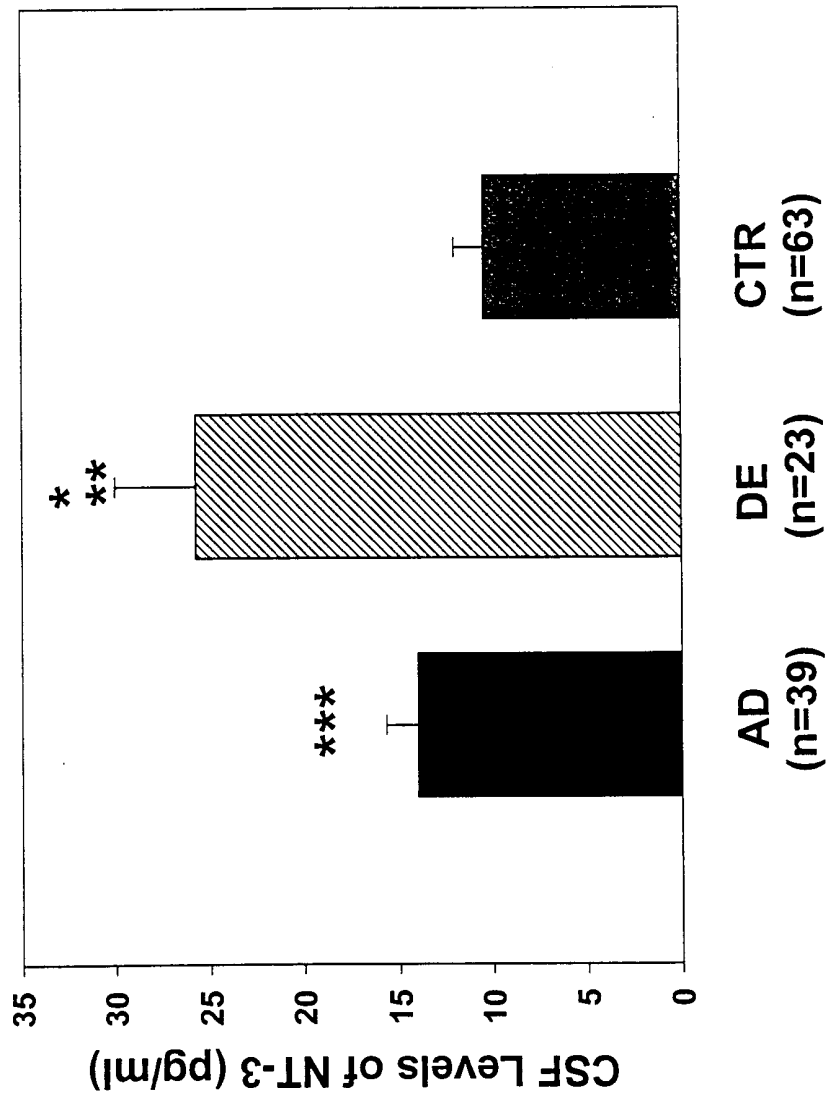


Fig. 3

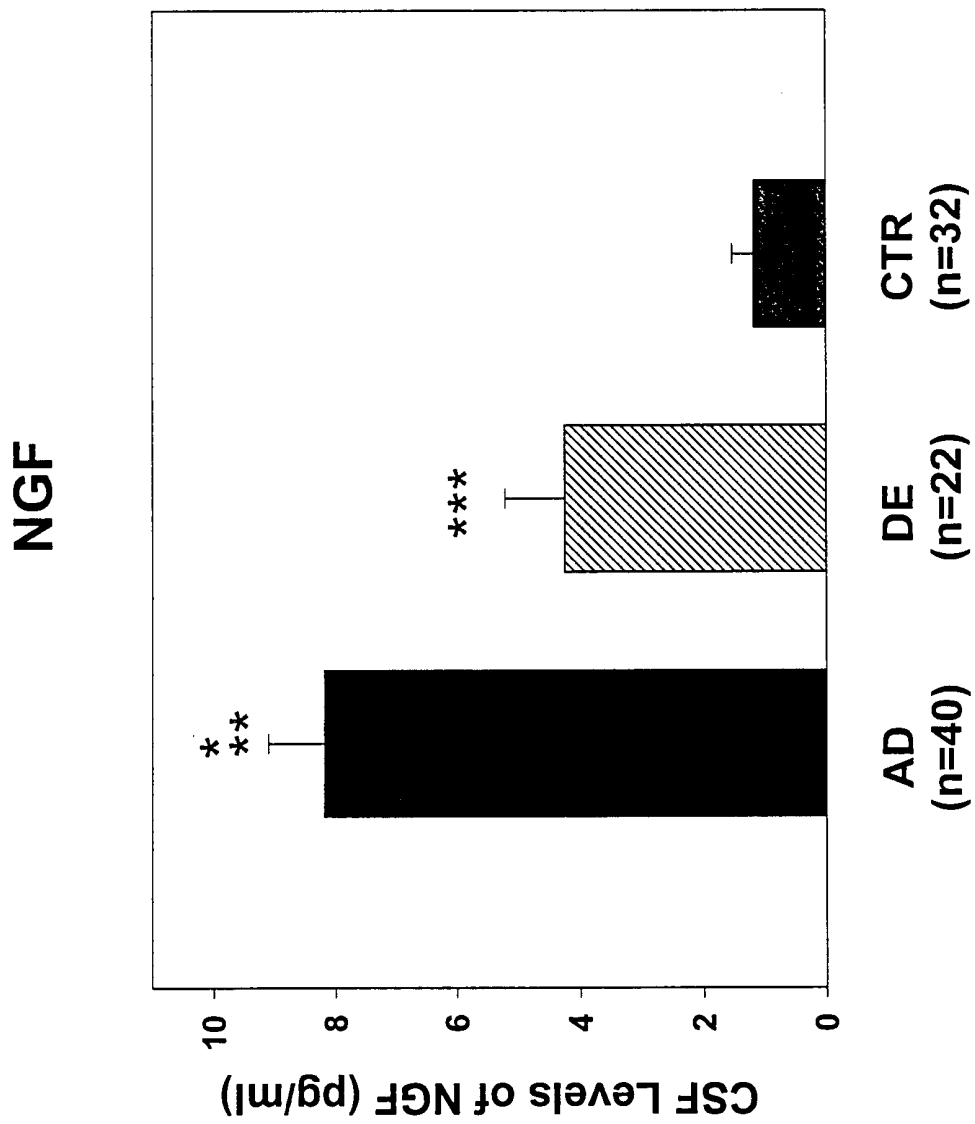


Fig. 4

Spinal Fluid Measurements of NGF and NT-3: Diagnostic Accuracy in Alzheimer's Disease (AD) and Major Depression in the Elderly (DE)

	NGF test (NGF>4 pg/ml)	NT-3 test (NT-3>15 pg/ml)
	Diagnosis of AD	Diagnosis of DE
Sensitivity	71,9%	73,9%
Specificity	79,2%	86,1%
Positive Predictive Value (PPV)	67,6%	50,0%
Negative Predictive Value (NPV)	82,4%	91,7%
	Combined NGF/NT-3 test (NGF>4 pg/ml, NT-3<15 pg/ml)	Combined NGF/NT-3 test (NT-3>15 pg/ml, NGF<4 pg/ml)
	Diagnosis of AD	Diagnosis of DE
Sensitivity	62.5%	55.5%
Specificity	90.1%	89.7%
Positive Predictive Value (PPV)	83.3%	71.4%
Negative Predictive Value (NPV)	76.9%	81.4%

Table 1

CSF Levels of NT-3 Grouped According to Treatment with Antidepressants

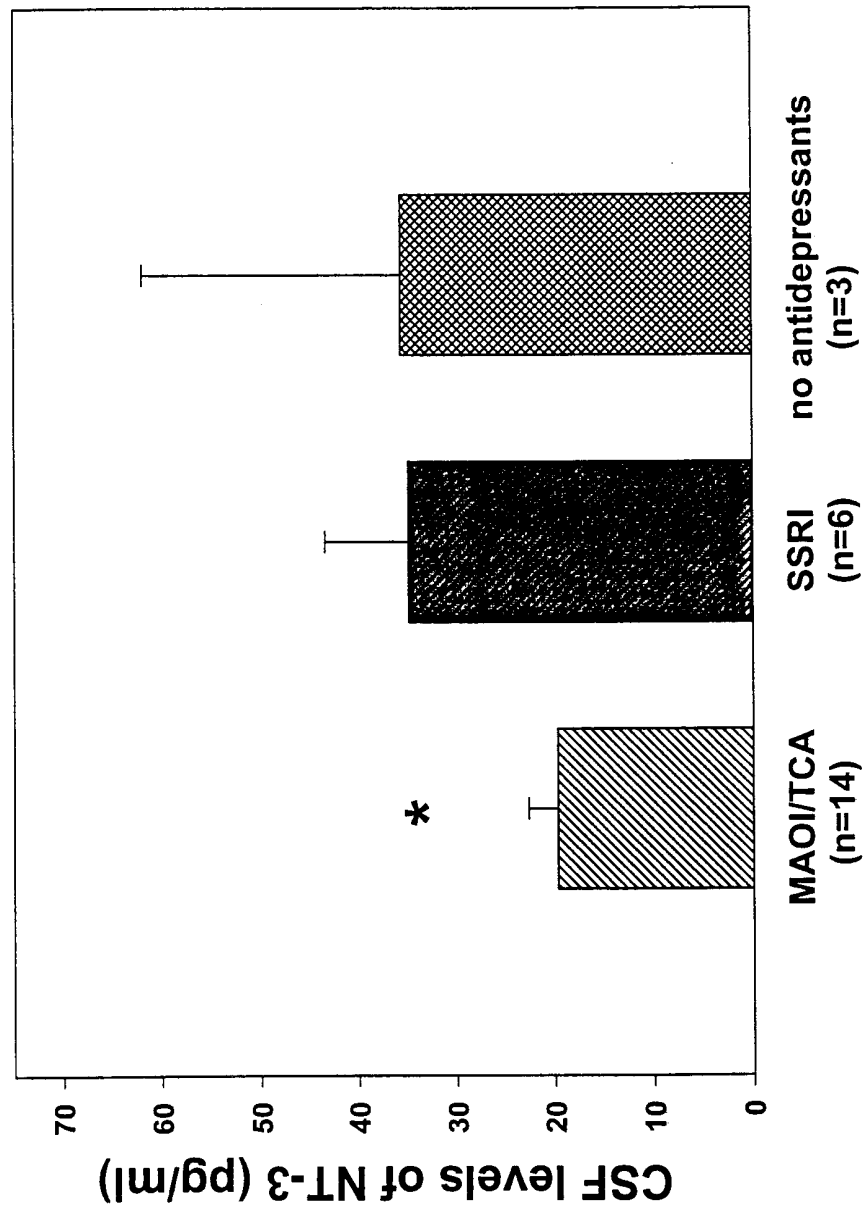


Fig. 5

INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 00/03912

A. CLASSIFICATION OF SUBJECT MATTER IPC 7 G01N33/68 A61K38/18				
According to International Patent Classification (IPC) or to both national classification and IPC				
B. FIELDS SEARCHED				
Minimum documentation searched (classification system followed by classification symbols) IPC 7 G01N C07K A61K				
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched				
Electronic data base consulted during the international search (name of data base and, where practical, search terms used) EPO-Internal, WPI Data, MEDLINE, BIOSIS, CHEM ABS Data				
C. DOCUMENTS CONSIDERED TO BE RELEVANT				
Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.		
X	WO 91 03569 A (MAX PLANCK GESELLSCHAFT ;REGENERON PHARMA (US)) 21 March 1991 (1991-03-21) cited in the application abstract page 38, line 29 -page 44, line 12 claims 82-128,137-140 ---	1-21		
X	MURASE, K.; IGARASHI, K.; HAYASHI, K.: "Neurotrophin-3 (NT-3) levels in the developing rat nervous system and in human samples" CLINICA CHIMICA ACTA, vol. 227, no. 1-2, 1994, pages 23-36, XP000852953 cited in the application abstract ---	1-18		
--- -/--				
<input checked="" type="checkbox"/> Further documents are listed in the continuation of box C.				
<input checked="" type="checkbox"/> Patent family members are listed in annex.				
° Special categories of cited documents :				
<table style="width: 100%; border: none;"> <tr> <td style="width: 50%; border: none; vertical-align: top;"> "A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed </td> <td style="width: 50%; border: none; vertical-align: top;"> "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art. "&" document member of the same patent family </td> </tr> </table>			"A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art. "&" document member of the same patent family
"A" document defining the general state of the art which is not considered to be of particular relevance "E" earlier document but published on or after the international filing date "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified) "O" document referring to an oral disclosure, use, exhibition or other means "P" document published prior to the international filing date but later than the priority date claimed	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art. "&" document member of the same patent family			
Date of the actual completion of the international search <p style="text-align: center; font-size: 1.2em;">28 September 2000</p>		Date of mailing of the international search report <p style="text-align: center; font-size: 1.2em;">06/10/2000</p>		
Name and mailing address of the ISA European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016		Authorized officer <p style="text-align: center; font-size: 1.2em;">Hart-Davis, J</p>		

INTERNATIONAL SEARCH REPORT

International Application No
PCT/EP 00/03912

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT		
Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	NARISAWA-SAITO M; WAKABAYASHI K; TSUJI S; TAKAHASHI H; NAWA H: "Regional specificity of alterations in NGF, BDNF and NT-3 levels in Alzheimer's disease" NEUROREPORT, vol. 7, no. 18, 25 November 1996 (1996-11-25), pages 2925-2928, XP000852945 cited in the application figure 2	1-18
X	WO 93 09798 A (REGENERON PHARMA) 27 May 1993 (1993-05-27) cited in the application claims 1,11-15	19-21
A	M BOTHWELL: "Functional interactions of neurotrophins and neurotrophin receptors" ANNUAL REVIEW OF NEUROSCIENCE, vol. 18, 1995, pages 223-253, XP000852678 cited in the application the whole document	1-20
A	GILMORE JH, JARSKOG LF, LINDGREN JC, MCEVOY JP, XIAO H: "Neurotrophin-3 levels in the cerebrospinal fluid of patients with schizophrenia or medical illness" PSYCHIATRY RESEARCH, vol. 63, no. 1-2, 14 November 1997 (1997-11-14), pages 109-113, XP000852677 cited in the application the whole document	1-20
A	E ARENAS, H PERSSON: "Neurotrophin-3 prevents the death of adult central noradrenergic neurons in vivo" NATURE, vol. 367, 27 January 1994 (1994-01-27), pages 368-371, XP002122262 cited in the application the whole document	19,20
A	M A SMITH, S MAKINO, M ALTEMUS, D MICHELSON, S-K HONG, R KVETNANSKY, R M POST: "Stress and antidepressants differentially regulate neurotrophin 3 mRNA expression in the locus coeruleus" PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES OF USA, vol. 92, September 1995 (1995-09), pages 8788-8792, XP002122263 cited in the application the whole document	1-20
	-/--	

INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 00/03912

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT		
Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>EBADI, M.; BASHIR, R. M.; HEIDRICK, M. L.; HAMADA, F. M.; EL REFAEY, H.; HAMED, A.; HELAL, G.; BAXI, M. D. ET AL.: "Neurotrophins and their receptors in nerve injury and repair" NEUROCHEMISTRY INTERNATIONAL, vol. 30, no. 4/5, 1997, pages 347-374, XP000852951 the whole document</p> <p style="text-align: center;">---</p>	19,20
A	<p>LEONARD BE: "The role of noradrenaline in depression: a review" JOURNAL OF PSYCHOPHARMACOLOGY, vol. 11, no. 4S, 1997, pages 39-47, XP000852762 cited in the application the whole document</p> <p style="text-align: center;">-----</p>	1-20

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 19-20 (partially)

Present claims 19-20 relate to an agent defined by reference to a desirable property, namely its ability to affect the activity and/or level of neurotrophin 3 (NT-3) or a gene coding for NT-3.

No technical features of the agents are present in the above-mentioned claims which would lead to this desirable property, the technical features formulated so as to permit the execution of a meaningful search. No support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is to be found for the substances which could fall within the scope of these claims. In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible. No means are present in the above-mentioned claims by which agents known in the prior art could be distinguished from novel agents. No definition of the subject matter for which protection is sought is therefore derivable from these claims (Article 6 PCT) or the description (Article 5 PCT). Independent of the above reasoning, the claims also lack clarity (Article 6 PCT). An attempt is made to define the compound by reference to a result to be achieved. Again, this lack of clarity in the present case is such as to render a meaningful search over the whole of the claimed scope impossible.

Consequently, the search for claims 19-20 has been restricted to the substances which are clearly defined and supported by the description, namely neurotrophin 3 (NT-3), in so far as it achieves the result of modulating the level of neurotrophin 3 (NT-3).

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 1-20

Diagnosis or monitoring or treatment or prevention of depression involving neurotrophin 3.

2. Claim : 21

Method of screening for agents influencing the activity and/or level of neurotrophin 3 or a gene coding for neurotrophin 3.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP 00/03912

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9103569 A	21-03-1991	US 5180820 A	19-01-1993
		AT 148921 T	15-02-1997
		AT 153074 T	15-05-1997
		AU 647412 B	24-03-1994
		AU 6337390 A	08-04-1991
		AU 643705 B	25-11-1993
		AU 6404990 A	08-04-1991
		CA 2040412 A	01-03-1991
		CA 2040437 A	01-03-1991
		CN 1052141 A	12-06-1991
		CN 1052142 A	12-06-1991
		CZ 9004229 A	16-06-1999
		CZ 286015 B	15-12-1999
		CZ 9004230 A	15-09-1999
		DD 299196 A	02-04-1992
		DD 299197 A	02-04-1992
		DE 69029934 D	27-03-1997
		DE 69029934 T	24-07-1997
		DE 69030719 D	19-06-1997
		DE 69030719 T	25-09-1997
		DK 440777 T	21-04-1997
		EP 0440777 A	14-08-1991
		EP 0441947 A	21-08-1991
		ES 2098271 T	01-05-1997
		ES 2100891 T	01-07-1997
		GR 90100647 A, B	30-12-1991
		GR 90100653 A, B	30-12-1991
		HK 1006578 A	05-03-1999
		HK 1006579 A	05-03-1999
		JP 2783450 B	06-08-1998
		JP 5328974 A	14-12-1993
		JP 5161493 A	29-06-1993
		KR 188189 B	01-06-1999
		KR 196203 B	15-06-1999
		LT 1546 A, B	25-07-1995
		LT 1818 A, B	25-09-1995
		LV 10725 A	20-06-1995
		LV 10725 B	20-12-1995
		LV 10792 A	20-08-1995
		LV 10792 B	20-12-1995
		NO 303694 B	17-08-1998
		NO 303584 B	03-08-1998
		NZ 235086 A	26-10-1993
		PT 95152 A, B	22-05-1991
		PT 95153 A, B	22-05-1991
		SG 46954 A	20-03-1998
		SK 422990 A	11-02-1999
		SK 423090 A	11-02-1999
WO 9309798 A	27-05-1993	AU 3130493 A	15-06-1993
		CN 1073264 A	16-06-1993
		PT 101051 A	28-02-1994
		ZA 9208730 A	10-05-1993