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Neurodegeneration Markers in the Cerebrospinal Fluid of 100 Patients with Schizophrenia Spectrum Disorder

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***Background*: Schizophrenia spectrum disorders (SSD) can be associated with neurodegenerative processes causing disruption of neuronal, synaptic, or axonal integrity. Some previous studies have reported alterations of neurodegenerative markers (such as amyloid beta [Aβ], tau, or neurofilaments) in patients with SSD. However, the current state of research remains inconclusive. Therefore, the rationale of this study was to investigate established neurodegenerative markers in the cerebrospinal fluid (CSF) of a large group of patients with SSD. *Study Design*: Measurements of Aβ1–40, Aß1–42, phospho- and total-tau in addition to neurofilament light (NFL), medium (NFM), and heavy (NFH) chains were performed in the CSF of 100 patients with SSD (60 F, 40 M; age 33.7 ± 12.0) and 39 controls with idiopathic intracranial hypertension (33 F, 6 M; age 34.6 ± 12.0) using enzyme-linked immunoassays. *Study Results*: The NFM levels were significantly increased in SSD patients (P = .009), whereas phospho-tau levels were lower in comparison to the control group (P = .018). No other significant differences in total-tau, beta-amyloid-quotient (Aβ1–42/Aβ1–40), NFL, and NFH were identified. *Conclusions*: The findings argue against a general tauopathy or amyloid pathology in patients with SSD. However, high levels of NFM, which has been linked to regulatory functions in dopaminergic neurotransmission, were associated with SSD. Therefore, NFM could be a promising candidate for further research on SSD.**

Key words: NFM/psychosis/SSD/neurofilament/tau/amyloid

Introduction

Schizophrenia is a common and severe mental illness characterized by positive (such as delusions and hallucinations) and negative symptoms (such as diminished emotional expression and cognitive impairment).[1] However, despite intensive research, the exact pathomechanism of the disease remains unclear. Many authors have interpreted schizophrenia as a neurodevelopmental disorder in which genetic and environmental factors have a strong influence.[2] Accordingly, prenatal, perinatal, childhood, or adolescent brain alterations can contribute to the development of schizophrenia, based on findings of reduced cortical volume, altered gyrification patterns, and ventricular enlargement at disease onset.[3] Another prominent neurodevelopmental theory suggests that risk factors in early adolescence can result in dysfunctional brain maturation during synaptic reorganization and pruning.[4] Nevertheless, these models cannot explain why schizophrenia generally does not present clinically until a decade after the cortex reaches peak cortical thickness and cortical restructuring has started.[3]

Over 100 years ago, Kraepelin discussed schizophrenia in the context of neurodegenerative disease as "dementia praecox," referring to the early onset of cognitive decline that slowly deteriorates with age.[5] Currently, evidence from imaging, postmortem, and biomarker studies suggests that neurodegenerative models (involving the disruption of neurons, synapses, or axonal integrity) might complement neurodevelopmental models.[3,6,7] For example, patients with schizophrenia display similar white matter lesions on magnetic resonance imaging (MRI) as patients with Alzheimer's dementia.[8] The examination

of cerebrospinal fluid (CSF) is particularly useful for biomarker studies due to its direct contact with brain structures. A large number of CSF biomarkers that have already been established for dementia have also been investigated in patients with schizophrenia. For example amyloid beta (Aβ) peptides, which are decreased in CSF in Alzheimer's disease, revealed an altered profile in patients with schizophrenia compared to elderly controls.[9] The microtubule-associated protein tau, which is increased in Alzheimer's disease (and other dementias, such as supranuclear gaze palsy or Picks disease), was reported to be decreased in the serum of patients with schizophrenia.[10] Finally, neurodegeneration markers such as neurofilaments have been studied to differentiate dementia from primary psychiatric disorders (such as schizophrenia) when symptoms such as psychosis or depression overlap.[11–13]

Neurofilaments belong to the category of intermediate filaments and form important structural scaffolding for the assembly of neurons.[14] Moreover, they are among the most dominant proteins in the brain and play an important role in nerve conduction by expanding the axonal caliber.[15,16] Unlike non-neuronal intermediate filaments, they are regulated by phosphorylation and form complex hetero-polymers containing four subunits with different molecular weights: neurofilament light (NFL), medium (NFM), and heavy (NFH) chains as well as α-internexin.[17] In addition, due to exclusive expression in neurons and the high measurable levels in CSF during neuroaxonal injuries and neuronal cell destruction, neurofilaments are a potentially valuable biomarker for a wide variety of neurological disorders that incorporate neurodegenerative, inflammatory, and traumatic processes.[14] However, in addition to alterations in neurological diseases (including multiple sclerosis, dementia, strokes, and Parkinson's disease), some alterations have also been found in mental disorders (such as bipolar disorders and schizophrenia).[6,14,18,19] A genetic link to schizophrenia has already been established, as the chromosomal regions of NFL, NFM, and NFH are associated with schizophrenia.[20,21] This is relevant to mental disorders because, in addition to their structural function and enhancement of signal transduction, neurofilaments may also exert other functions that involve the regulation of synapses and neurotransmission.[15,17,22]

Accordingly, the aim of this study was to investigate these neurodegenerative markers, which are well established for mainly neurologic diseases, in the CSF of a large group of patients with schizophrenia spectrum disorders (SSD; for previous findings, see table 1).

Methods

Ethical approval for this study was granted by the Ethics Committee of the University of Freiburg in the context of a larger retrospective research project (EK-Fr 609/14). Written informed consent was obtained from all patients with SSDs before lumbar puncture. All neurological control patients were contacted retrospectively and asked to supply written informed consent to use their residual serum/CSF material for research purposes.

Study Sample

For this study, a previously published retrospective group of 100 patients with SSD (60 females, 40 males; mean age with standard deviation: 33.72 ± 12.05 years) was used (see table 2).[25] All patients with SSD were above 18 years of age and received a lumbar puncture as part of the routine diagnostic workup. Diagnoses were established by experienced psychiatrists according to ICD-10 criteria upon hospital admission. Patients with known substance use and immunological disorders known for brain involvement were excluded. All patients of the study group received an MRI scan of the brain and an electroencephalography (EEG). For the majority of patients, comprehensive demographic and clinical data and psychometric scales, such as the Global Assessment of Functioning (GAF), Clinical Global Impression (CGI), and psychopathological scores based on guidelines published by the German Association for Methodology and Documentation in Psychiatry (AMDP) were obtained as part of the admission routine.

As a control group, another previously established group of 39 patients (33 females, 6 males; mean age with standard deviation: 34.62 ± 12.03 years) with idiopathic intracranial hypertension (IIH) was analyzed.[26–29] IIH is a noninflammatory CNS disease, that requires often repeated lumbar punctures with CSF withdrawal for diagnosis and treatment. All IIH controls had no known psychiatric diagnosis and took no psychotropic medication.

No significant age difference ($z = -0.516$, $P = .606$) was observed between SSD patients and controls. However, a significant sex difference ($\chi^2 = 7.678$, $P = .006$) between the two predominantly female groups was detected. The SSD group mainly comprised patients with paranoid schizophrenia (56%) or schizoaffective disorder (30%). Moreover, chronic or recurrent courses (58%) were included more often than first-time diagnoses (42%). All clinical and demographic data are presented in more detail in an earlier publication of the patient group.[25]

Cerebrospinal Fluid Analysis

All routine CSF analyses were conducted in the CSF laboratory of the Department of Neurology and Neurophysiology at the Medical Center of the University of Freiburg. CSF and serum not used in routine analysis were aliquoted and stored at −80°C for future analysis. All CSF diagnostics are described in more detail in previous publications.[26,27,30,31]

Table 1. Selected Studies Investigating Neurodegenerative Markers in Patients With Schizophrenia Spectrum Disorders

	Cohorts	Average Age in Years ± SD, Age Range, (% of males)	Neurodegenerative Markers Measured (Method)	Results
Frisoni et al. 2011,[23]	$N = 11$ elderly schizophrenia patients	69.2 ± 2.0 (27)	Aβ1–42, tau, and phospho-tau in CSF (ELISA)	Significant reduction of Aβ1–42 levels of patients with schizophrenia compared to controls. Lower tau and phospho-tau were not statistically significant. In Alzheimer's disease Aβ1–42 levels were significantly decreased and tau levels significantly increased compared to the other two groups. Strong reduction of almost all Aβ species in CSF from schizophrenia group, while in Alzheimer's disease only Aβ1–42 was reduced
Albertini et al. 2012[9]	$N = 20$ sporadic Alzheimer's disease	70.6 ± 1.7 (40)	15Aβ isoforms in CSF (SELDI TOF mass spectrometry)	
	$N = 20$ cognitively healthy age-matched controls	65.3 ± 1.9 (70)		
Andreou et al. 2021[10]	$N = 37$ early-onset psychosis	16.4 ± 1.3 (30)	Total-tau in plasma (SiMoA)	Significantly lower plasma tau levels in early-onset psychosis compared to controls
	$N = 20$ healthy age-matched controls	16.2 ± 1.5 (46)		
Rodrigues-Amorim et al. 2020[6]	$N = 42$ schizophrenia	41.10 ± 14.41 (62)	Plasma β-III tubulin, neurofilament light chain, and glial fibrillary acidic protein (quantitative immunoblotting)	Significantly elevated plasma levels of all three proteins in schizophrenia patients compared to controls
	$N = 40$ healthy age-matched controls	44.96 ± 14.95 (63)		
Al Shweiki et al. 2019[12]	$N = 11$ schizophrenia patients (6 with paranoid and 5 undifferentiated subtypes)	41.1 (45)	Neurofilament light chain in plasma (SiMoA)	No statistically significant difference in plasma neurofilament light chain levels between schizophrenia patients and controls
	$N = 27$ controls	46.8 (7)		
Guasp et al. 2022[24]	$N = 45$ first episode psychosis (35 schizophrenia and 10 bipolar)	20 (56)	Neurofilament light chain in plasma (SiMoA)	Significantly higher neurofilament light chain in plasma of patients with NMDA-R encephalitis (27.5 pg/ml) compared to patients with psychosis (7.1 pg/ml) and controls (5.1 pg/ml). The levels between patients with primary psychosis and healthy controls were not statistically compared/reported
	$N = 118$ anti-NMDA-R encephalitis	23 (17)		
	$N = 36$ healthy controls	25 (42)		

Note: CSF, cerebrospinal fluid; *N*, number; NMDA-R, N-methyl-D-aspartate receptor; SD, standard deviation; SiMoA, Single molecule array.

Neurodegenerative Markers

To determine the neurodegenerative markers, the CSF of the patients and controls was analyzed with commercially available enzyme-linked immunosorbent assays (ELISAs). The assays for phospho-tau (phosphorylation at threonine 181), total-tau, and amyloid (1–40 and 1–42) were developed for measurements in CSF and used according to the manufacturer's instructions (Euroimmun, Lübeck, Germany). It should be noted that CSF samples were collected in polystyrene tubes, which can cause aggregation of amyloid proteins and therefore false low values.[32–34] However, given that amyloid 1–42 and 1–40 are affected in similar proportions, the amyloid quotient (Aβ1–42/Aβ1–40) remains valid.[34–37] Therefore, for amyloid-β proteins 1–42 and 1–40, only the quotient is reported.[34] For this study, clinically common reference values for total-tau (<450 pg/ml), phospho-tau (<61 pg/ml), and the beta-amyloid-quotient (>0.05) were used. For the findings of the NFL protein (Tecan Trading AG, Switzerland), NFM (Cusabio Technology LLC, Houston, USA), and phosphorylated NFH chains (Euroimmun, Lübeck, Germany), only group comparisons were performed. Moreover, assays that could only detect concentration values for <50% of the samples were not further analyzed. This was only the case for phosphorylated NFH chains, which could only be measured in less than 33% of the patients.

Statistical Analyses

All acquired data were collected and analyzed using SPSS Version 28 (IBM, Armonk, USA) and R.[38] Any differences in categorical variables (such as sex or number of participants with pathological neurodegenerative markers) between SSD patients and controls, as well as in between SSD subgroups, were analyzed by Chi-squared tests. For age comparisons, a nonparametric Mann–Whitney *U* test was employed. For all neurodegeneration markers, we checked whether the requirements for parametric analyses of covariance (ANCOVA) were met.

Table 2. Clinical Data of Patients With Schizophrenia Spectrum Disorder

	SSD Patients ($N = 100$)
Diagnoses	
Paranoid schizophrenia [F20.0]	56 (56%)
Hebephrenic schizophrenia [F20.1]	2 (2%)
Catatonic schizophrenia [F20.2]	2 (2%)
Delusional Disorder [F22.0]	6 (6%)
Acute polymorphic psychotic [F23.1]	4 (4%)
Schizoaffective disorder [F25.X]	30 (30%)
Course of disease	
First-time diagnosis	42 (42%)
Chronic/recurrent	58 (58%)
Psychotropic drugs at the time of sampling	
SSRI	7(7%)
SSNRI	5 (5%)
Mirtazapine	3 (3%)
Tricyclic antidepressants	2 (2%)
Typical antipsychotics with high-potency	6 (6%)
Typical antipsychotics with low-potency	10 (10%)
Atypical antipsychotics	96 (96%)
Lithium	10 (10%)
Benzodiazepines	16 (16%)
Anticonvulsants	15 (15%)
Unmedicated	3 (3%)
Number of previous inpatient stays	
None	35 (35%)
1	12 (12%)
2	10 (10%)
3	9 (9%)
More than 3	20 (20%)
Unknown	14 (14%)
Number of suicide attempts	
None	65 (65%)
1	8 (8%)
2	9 (9%)
More than 2	3 (3%)
Unknown	15 (15%)

Note: SSRI, selective serotonin reuptake inhibitor; SSNRI, selective serotonin/noradrenaline reuptake inhibitor.
Extract of published data in Runge et al. 2022.[25]

Their normality of distribution was tested in the corresponding study groups using the Shapiro–Wilk test, and the homogeneity of variance between groups was tested using Levene's test (see Supplemental Table S1). Because of multiple violations of normality and homogeneity of variance, all statistical differences in neurodegenerative markers between groups were assessed using nonparametric analysis of covariance (ANCOVA), with sex as a covariate to control for the significant sex differences, using raov function of R package rfit.[38,39] To minimize false discovery rates in the main neurodegeneration analysis, the Benjamini–Hochberg procedure for multiple comparisons was performed with R.[38] Furthermore, subgroup analyses between paranoid-hallucinatory ($n = 56$) and schizoaffective patients ($n = 30$), as well as against the control group ($n = 39$), were conducted with the same ANCOVA model. Finally, SSD patients with chronic course ($n = 58$) were compared against first-time diagnosis ($n = 42$) in this manner.

Moreover, correlation analyses of neurodegenerative markers with age, CSF routine parameters, psychometric scales, and demographic data of the SSD patients were performed with nonparametric Spearman's rank correlation coefficient. For a secondary analysis, when investigating the influence of blood–brain barrier dysfunction on neurofilament concentrations, elevated/nonelevated albumin quotient was used in addition to study group and sex as a further group factor in the nonparametric ANCOVA model. Due to the exploratory approach, no correction for multiple testing was applied in the correlation and subgroup analyses. For all statistical analyses, a significance level of <0.05 was defined, and all plots were created with the R package ggplot2[38,40] and Adobe Illustrator (Adobe Inc., San José, CA).

Results

Neurodegenerative Markers

In the CSF of patients and controls, the concentrations of total-tau, phospho-tau, amyloid β1–42, amyloid β1–40, NFL, and NFM chains were adequately measurable.

In the control group of patients with IIH, significantly higher levels of phospho-tau ($P = .018$) were detected. Nevertheless, only one control patient exhibited a minimally pathological total-tau value of 457.69 pg/ml, and no patients with pathological phospho-tau levels were observed. One female SSD patient with a reduced beta-amyloid-quotient of 0.026 showed no other pathological neurodegenerative markers. The NFM concentrations were significantly higher in the patients with SSD ($P = .009$).

In these models, no significant influence of sex on neurodegenerative markers could be found. However, a significant interaction between the study groups and sex was observed for NFM ($F_{(1,133)} = 4.453$, $P = .037$). The results of the neurodegenerative marker levels are presented in figure 1 and table 3.

Subgroup Analyses

Subgroup analyses were conducted to compare patients with paranoid-hallucinatory and schizoaffective syndromes with the control group (and between themselves), as well as between SSD patients with chronic course and first-time diagnoses. Between the different subgroups, no significant differences in age were found. For sex, only differences between SSD patients with paranoid-hallucinatory syndrome and controls ($\chi^2 = 9.918$, $P = .002$) were detected.

Hereby, differences between phospho-tau stayed significant for paranoid-hallucinatory syndromes ($F_{(1,91)} = 6.341$, $P = .014$), but not schizoaffective syndromes ($F_{(1,65)} = 2.293$, $P = .135$). For NFM both syndromes remained

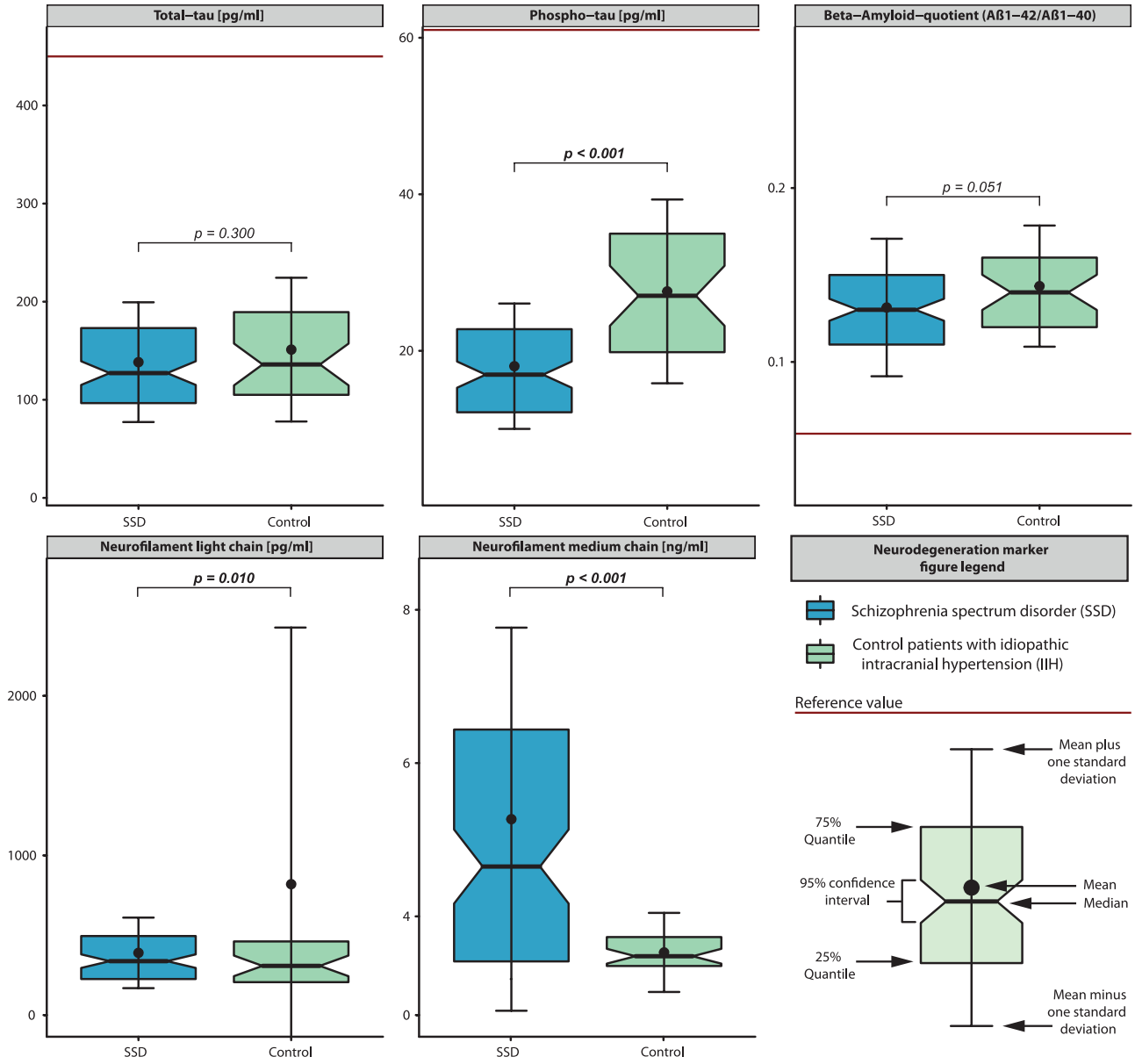

Fig. 1. Boxplot of neurodegenerative markers in cerebrospinal fluid of patients with schizophrenia spectrum disorders (SSD) and controls. Reference values for total-tau, phospho-tau, and beta-amyloid-quotient are indicated as thin horizontal lines. Error bar of the neurofilament light chain is not completely shown for the control group. The difference between mean and median is caused by a few extreme outliers in the control group.

statistically significant (paranoid-hallucinatory: $F_{(1,90)}$ = 14.114, $P < .001$; schizoaffective: $F_{(1,63)} = 7.313$, $P = .009$). When comparing patients with paranoid-hallucinatory and schizoaffective syndromes or chronic course and first diagnosis of the disorder, no concentration differences in neurodegenerative markers were observed.

Correlation Analyses

NFL correlated with phospho-tau ($r = 0.493$, $P < .001$, $n = 100$), total-tau ($r = 0.463$, $P < .001$, $n = 100$), NFM ($r = 0.324$, $P = .001$, $n = 98$), and age ($r = 0.527$, $P < .001$, $n = 100$). Besides NFL, NFM was also correlated with phospho-tau ($r = 0.217$, $P = .032$, $n = 98$), total-tau ($r = 0.343$, $P < .001$, $n = 98$), and age ($r = 0.277$, $P = .006$, $n = 98$), as well as phospho-tau with total-tau ($r = 0.741$, $P < .001$, $n = 100$), and age ($r = 0.205$, $P = .040$, $n = 100$). Beta-amyloid quotients correlated negatively with total-tau ($r = -0.287$, $P = .004$, $n = 100$) and the psychometric scores for orientation disturbances ($r = 0.270$, $P = .016$, $n = 79$), disorders of perception ($r = 0.236$, $p = .043$, $n = 74$), and formal thought disorder ($r = 0.280$, $P = .012$, $n = 80$). The latter also correlated with NFL ($r = 0.232$, $P = .039$, $n = 80$). Further correlations of psychometric scales with neurodegenerative markers could not be observed.

Table 3. Neurodegenerative Markers in CSF of Patients With Schizophrenia Spectrum Disorders and Controls

	Patients With Schizophrenia Spectrum Disorders ($n = 100$)	Controls ($n = 39$)	Statistics[b]
Total-tau [pg/ml]	138.33 ± 61.04	151.14 ± 73.27	$F_{(1,136)} = 0.004$, $P = .949$
Phospho-tau [pg/ml]	18.03 ± 8.01	27.59 ± 11.74	$F_{(1,136)} = 7.495$, **$P = .018$**
Beta-amyloid-quotient (Aβ1-42/Aβ1-40)	0.131 ± 0.039	0.142 ± 0.035	$F_{(1,136)} = 2.092$, $P = .251$
Neurofilament light chain [pg/ml]	389.26 ± 220.92	819.51 ± 1607.84	$F_{(1,136)} = 0.057$, $P = .949$
Neurofilament medium chain[a] [ng/ml]	5.27 ± 2.50 ($n = 98$)	3.53 ± 0.52 ($n = 38$)	$F_{(1,133)} = 10.234$, **$P = .009$**

Note: Values ± standard deviation. Significant *P*-Values are indicated in bold.
[a]Not measurable in three subjects.
[b]*P*-value adjusted for multiple testing.

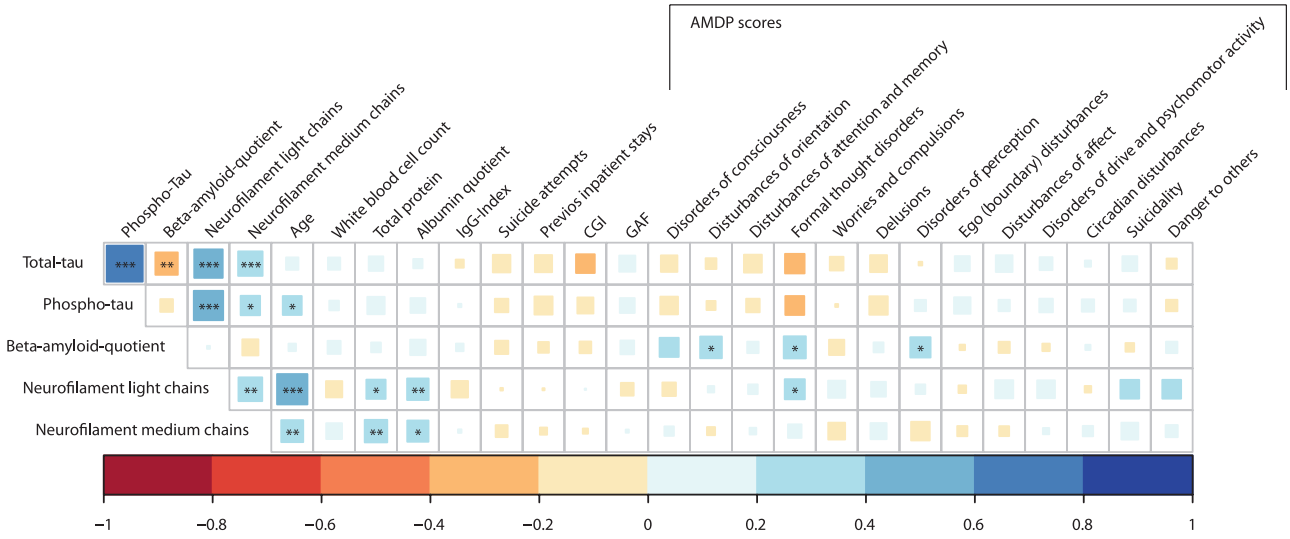

Fig. 2. Correlation of neurodegenerative markers with age, cerebrospinal fluid routine parameters, psychometric scales, and demographic data. Correlations were performed with Spearman rho correlation coefficients. Significant results are marked with asterisks (* represents a *P*-value below .05, ** below .01, and *** below .001). Abbreviations: AMDP, German Association for Methodology and Documentation in Psychiatry; CGI, Clinical Global Impression; GAF, Global Assessment of Functioning; IgG, Immunoglobulin G.

Regarding basic CSF parameters, significant correlations between the neurofilaments and total protein (NFL: $r = 0.213$, $P = .034$, $n = 100$; NFM: $r = 0.292$, $P = .004$, $n = 98$) as well as the albumin quotient (NFL: $r = 0.266$, $P = .007$, $n = 100$; NFM: $r = 0.259$, $P = .010$, $n = 98$) were detected. Given the correlation between neurofilaments and albumin quotient as a parameter for blood–brain barrier function, we performed a secondary analysis with sex and elevated/nonelevated albumin quotient as covariates to exclude possible bias by blood-CSF barrier dysfunction. There was still a significant difference between the SSD and control groups for NFM ($F_{(1,128)} = 9.071$, $P = .003$). No significant effects of sex or elevated albumin quotient on neurofilament concentrations were observed in this model. No significant correlations of neurodegenerative markers with the number of suicide attempts or previous inpatient stays were observed (figure 2).

Discussion

The main finding of this study was the detection of higher NFM levels in the CSF of patients with SSD compared to the control group of patients with IIH. Furthermore, the IIH patients presented with higher phospho-tau concentrations, whereas the beta-amyloid quotients exhibited no group differences.

Neurofilament Medium and Schizophrenia

Together with NFH, NFM is an intermediate filament subunit with a very long C-terminal tail. In particular, NFM is important for the growth of the axonal diameter,

in which phosphorylations in the C-terminus play an important role.[16] In comparison to its shorter counterpart (NFL), which is often studied as a biomarker for neurodegeneration, there are significantly fewer previous studies available. However, there is evidence of multiple functionalities of NFM in neurotransmission and interactions with dopamine receptors, which is particularly relevant for possible pathomechanisms in SSD.[15,17] At the genetic level, in addition to the association of the chromosomal localization of the gene *NEFM* (which encodes for NFM) with schizophrenia, two single nucleotide polymorphisms in *NEFM* have also been associated with early responses to antipsychotic medication in acutely psychotic patients with schizophrenia.[41] At the molecular level, NFM appears to bind the intracellular loop of the D1 dopamine receptor (D1R), regulating the expression of D1R on the cell surface and the corresponding sensitivity to D1R agonists.[42–44] It is believed that NFM-mediated anchoring of D1R in the endocytic-recycling compartment occurs shortly after receptor activation, meaning that the receptor is not promptly available again at the plasma membrane.[15] Moreover, a lack of NFM can result in increased sensitivity to D1R agonists through increased receptor density at the postsynaptic plasma membrane. This result is based on observations of a mouse model in which genetic deletion of NFM enhanced a D1R-mediated motor response caused by cocaine, with a higher accumulation of postsynaptic D1R at the plasma membrane.[15] This effect is found in the CNS and also in the adrenal gland through inhibition of the D1R-mediated neuroendocrine secretion of aldosterone by NFM.[45] In the different functioning roles of NFM, phosphorylation seems to be important, as less phosphorylated NFM has been found near the synapse compared to the axon.[15] In summary, there appears to be a link between NFM and schizophrenia on a genetic level. Moreover, based on the dopamine hypothesis of SSD, a possible pathomechanism may be D1R hypersensitivity due to NFM deficiency or dysfunction.

Elevated Neurofilament Medium Concentrations in Cerebrospinal Fluid

Although the association between schizophrenia and NFM is plausible, the reason for the elevated NFM levels in the CSF of SSD patients revealed in this study is unclear. Moreover, little is known about the CSF levels of NFM in schizophrenia, other diseases, or healthy controls in general. To date, increased CSF levels of NFM have been reported for amyotrophic lateral sclerosis (ALS),[13] frontotemporal dementia,[46] and cerebral hemorrhage.[47] Elevated serum NFM levels have also been found in ALS[48] and traumatic brain injuries.[47] Martínez-Morillo et al. reported NFM CSF levels in patients with subarachnoid hemorrhage of 5.66 ng/ml (n = 8), which were similar to the SSD values of 5.27 ng/ml (n = 98) in patients of this study. However, these are lower than in cases of intracerebral hemorrhage (10.6 ng/ml; n = 7) and higher than in control individuals (0.23 ng/ml; n = 10).[47] Although CSF concentrations are not known for patients with schizophrenia, autoantibodies to NFM in serum have been reported.[44] Autoantibodies to NFM appear to be common in neurologic diseases, including high levels in multiple sclerosis[49] or ALS.[50] In contrast to the increased NFM concentrations in CSF, postmortem studies on the brains of patients with schizophrenia have reported a decreased expression of NFM in several brain regions, including the corpus callosum, thalamus, and anterior cingulate cortex.[51–53] However, with regard to the thalamus, one study reported an increase in NFM in schizophrenia patients.[54] It remains unclear how elevated NFM concentrations in CSF occur in the presence of mainly decreased expression in certain brain regions. One hypothesis is that regional immunological or degenerative processes result in increased cell death or damage to regions containing NFM (ie, axons and dendritic spines). However, a dysregulation of the phosphorylation of NFM may also result in an accumulation of dysfunctional filaments, which might be shed into the interstitium or CSF without any cell death or degeneration. In the context of current knowledge on schizophrenia and NFM, this would be especially reasonable for dendritic spines, where hypophosphorylation of NFM may be important to keep it in place.[55] For ALS and Alzheimer's dementia, dysregulated neurofilament phosphorylation with the accumulation of hyperphosphorylated forms has been described.[48,56–58]

Other Neurodegenerative Markers

The study revealed no significant differences of beta-amyloid quotients in the CSF of patients with SSD in comparison to the control group, which is consistent with the findings of a comprehensive meta-analysis of 14 studies investigating the association of beta-amyloid burden and schizophrenia.[2] While there were also no significant differences in total-tau, the concentrations of phospho-tau and NFL were significantly reduced in the CSF of patients with SSD compared to the IIH control group. Unfortunately, these results are difficult to interpret, as increased tau concentrations may occur as a result of IIH pathology caused by increased intracranial pressure.[59,60] In the literature, no significant differences in CSF total-tau and phospho-tau have been reported,[23,61] although significantly lower serum total-tau levels in schizophrenic patients compared to controls have been found.[10,62] For NFL, many psychiatric studies have focused on distinguishing SSD from dementia[12,63] or even NMDA receptor encephalitis.[24] Postmortem brain studies in patients with schizophrenia have revealed decreased expression of NFL in various brain regions (such as the dorsolateral prefrontal cortex and corpus callosum).[17,64]

No significant differences in NFL concentrations in the serum of patients with schizophrenia were reported compared to the control groups, which is in line with the CSF findings of this study.[12,65,66]

Limitations

Some limiting factors in the study such as the open and retrospective study design should be noted. Lumbar punctures were performed as a routine diagnostic examination to exclude organic causes, but not for study purposes. Furthermore, the influence of sex on the study results cannot be excluded due to the significant differences between the study groups. In previous studies, the influence of sex on neurodegenerative markers has remained predominantly unclear. For example, in an Alzheimer's dementia cohort, a difference in phospho-tau levels between males and females was found in carriers of the apolipoprotein E ε4 allele, whereas this was not the case in noncarriers.[67] Similarly, some studies have reported sex-related differences in NFL,[18] while others found no such difference.[68] For NFM, higher levels have been reported for males in serum, although not in CSF.[47] Given this unclear influence, statistical comparisons of neurodegeneration markers were corrected for sex. Hereby the authors found a significant interaction between sex and study groups for NFM, although sex alone had no significant effect on NFM concentrations. For all other neurodegenerative markers, sex did not influence the statistical models. Another important influencing factor could be the antipsychotic medication of SSD patients. Especially for NFM, this cannot be ruled out due to its influence on neurotransmission in the dopaminergic system. With regard to tau protein, it has been reported that some antipsychotics can reduce both tau levels and phosphorylation, which could explain the significantly lower phospho-tau levels compared to the control group in this study.[10] Additionally, the IIH pathology of the control group could have influenced the results due to elevated tau concentrations under increased intracranial pressure.[59] It must be acknowledged, that the study cohorts with their mean age around 34 years are relatively young and therefore unlikely to suffer from neurodegenerative changes yet. Technically, the study was limited in terms of the significance of the amyloid findings, as the use of polystyrene tubes could have caused the amyloid aggregation. Therefore, only the amyloid quotient was used for the group comparisons. It should also be mentioned that other immunoassays (such as single molecule arrays [SiMoA]) can now provide even more sensitive results, which would facilitate other measurements (eg, neurofilaments in blood serum).

Conclusion

In contrast to models of general neurodegeneration or amyloid pathology, our results of increased NFM levels in the CSF of patients with SSD suggest local processes of neural alteration or NFM dysregulation. Although further studies are required to elucidate the pathomechanism in more detail, NFM appears to be a promising biomarker candidate for SSD.

Supplementary Material

Supplementary material is available at https://academic.oup.com/schizophreniabulletin/.

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Authors' Contributions

KR wrote the paper. DE critically revised the first draft. KR, AB, BLF, LTvE, and DE organized the study and created the study design. AB and BLF performed the laboratory measurements. RD performed the CSF routine analyses. KR, AB, and SJM performed the statistical analyses. AB, BLF, SJM, KvZ, KN, RD, KD, and LTvE revised the manuscript critically focusing on clinical and statistical aspects. All authors were critically involved in the theoretical discussion and composition of the manuscript. All authors read and approved the final version of the manuscript.

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