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RESEARCH ARTICLE

Increased oligomeric TDP-43 in the plasma of Korean frontotemporal dementia patients with semantic dementia

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Abstract

INTRODUCTION: Semantic dementia (SD) is a progressive neurodegenerative disease associated with impaired vocabulary that progresses to memory impairment. Post-mortem immunohistochemical analysis is the current reliable method of differentiating TDP-43 deposits in cortical tissue; no means of antemortem diagnosis exists in biofluids, let alone in plasma.

METHODS: Here the multimer detection system (MDS) was used to quantify the oligomeric TDP-43 (o-TDP-43) concentrations in plasma of Korean SD patients ($n = 16$, 6 male, 10 female, ages 59–87). The o-TDP-43 concentrations were compared with the total TDP-43 (t-TDP-43) concentrations quantified through conventional enzyme-linked immunosorbent assay (ELISA).

RESULTS AND DISCUSSION: Only MDS showed a significant increase in o-TDP-43 concentrations in the plasma of patients with SD compared to other neurodegenerative disorders and normal controls ($p < 0.05$). Based on these results, o-TDP-43 concentrations through the application of MDS may be a useful plasma biomarker in SD-FTD (frontotemporal dementia) diagnosis.

KEYWORDS
aggregation, antibodies, diagnosis, elisa, epitope, immunoassay, monomer, multimer detection system, oligomer, semantic dementia, TDP-43

1 | BACKGROUND

Semantic dementia (SD) is described as memory loss that primarily affects skills related to language and retention of vocabulary.[1,2] Neuronal degeneration primarily starts out at the anterior temporal lobes and temporal pole and may include the hippocampus and amygdala. The ventromedial prefrontal cortical regions may also be affected as the disease progresses.[3] SD is principally a phenotypic variant of frontotemporal dementia (FTD), which is a progressive neurodegenerative disease characterized mainly by the degeneration of the frontal and temporal lobes.[4] FTD has three clinical subtypes first described in 1998: (1) behavioral-variant FTD (bvFTD), indicated by

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abnormal changes in behavior and personality associated with frontal-pre-dominant cortical degeneration, (2) progressive non-fluent aphasia (PNFA), characterized by grammar and motor speech deficits associated with the left perisylvian cortical atrophy, and (3) semantic dementia (or SD), indicated by a gradual decline in the knowledge of words and word-object association linked to anterior temporal neuronal loss. Neuropathologically, FTD is classified into two types: FTD-TAU, characterized by tau pathology, and FTD-TDP, characterized by TDP-43 pathology.[4] However, the differences in the frequencies of tau and TDP-43 pathology cannot be directly translated to the clinical phenotypes particularly in PNFA cases.[4] Only SD presents a "pure" TDP-43 pathology.[5]

Increased total TDP-43 (t-TDP-43) concentrations were found previously in the plasma of 46% of FTD patients and 22% of Alzheimer's disease (AD) patients when compared to 8% of control subjects using capture monoclonal antibodies that bind to the RNA recognition motif (RRM-2) and detected by polyclonal antibodies specific to the N-terminal domain (NTD) of TDP-43.[6] The proportions of FTD and AD patients with increased TDP-43 levels in plasma were also found to correspond closely with the proportions of FTD and AD patients showing TDP-43 pathological changes in the brain based on previous autopsy studies. This was early evidence demonstrating that raised TDP-43 levels may index TDP-43 pathology within the brain.[6] TDP-43 may form oligomeric species (o-TDP-43) prior to aggregate formation.[7] In another study, an o-TDP-43–specific antibody was developed to discern TDP-43 oligomers from full-length TDP-43 through protein conformational differences in the frontal cortical and hippocampal sections of pathologically confirmed FTD-TDP subjects, three cases of age-matched non-dementia controls, and three cases of pathologically confirmed AD without TDP-43 pathology.[8] However, no other method for detecting o-TDP-43 in biological fluids has been described.

The multimer detection system (MDS) is an immunoassay-based approach of using capture and detection antibodies that bind to an overlapping epitope of a single antigen.[9] Monomers of the target protein are not detected in the MDS measurement because they only have one exposed epitope that is already obscured by the capture antibody, thus inhibiting the detection antibody from binding to that same epitope (Figure 1A). In contrast, oligomers present multiple but similar epitopes that allow the detection antibodies to attach while the protein is anchored by the capture antibodies (Figure 1B). This simple and effective approach was initially used to detect prion proteins in the plasma of scrapie-infected hamsters and was later expanded to detecting prion oligomers in sheep plasma as well as amyloid beta (Aβ) oligomers in plasma of patients with AD.[9–12]

In this study, o-TDP-43 and t-TDP-43 concentrations in the plasma of Korean SD-FTD patients were quantified using MDS and conventional sandwich enzyme-linked immunosorbent assay (ELISA), respectively, and the resulting concentrations were compared to o-TDP-43 and t-TDP-43 concentrations in plasma samples of Korean patients with AD, mild cognitive impairment (MCI), early-onset AD (EOAD), late-onset AD (LOAD), Parkinson's disease (PD), and age-matched healthy controls. In MDS, both the capture and detection polyclonal antibodies were specific for the NTD, whereas in the conventional sandwich ELISA, the capture antibody was NTD specific, whereas the detection antibodies were specific against the C-terminal domain (CTD). A significantly higher concentration of o-TDP-43 was found in the plasma of SD-FTD patients compared to the other groups. Expressing the o-TDP-43 concentration as a ratio with its corresponding t-TDP-43 concentration made the difference between SD-FTD and the other disease groups more distinct. In contrast, no significant differences in t-TDP-43 concentrations were found between the groups.

RESEARCH IN CONTEXT

1. **Systematic Review**: Publications on the detection, characterization, and quantification of TDP-43 oligomers in neurodegenerative diseases were reviewed. The deposition of TDP-43 is a well-established neurodegenerative marker, but the diagnostic potential of its oligomeric form is still being elucidated, particularly in semantic dementia.
2. **Interpretation**: Using a multimer detection system that differentiates oligomeric TDP-43 (o-TDP-43) from monomeric TDP-43, o-TDP-43 concentrations were found to be significantly higher in the plasma of frontotemporal dementia patients with semantic dementia (SD-FTD) compared to healthy controls and other disorders. This supports the diagnostic potential of o-TDP-43 in discriminating SD-FTD among other neurodegenerative diseases.
3. **Future Directions**: The ability of this approach to discriminate FTD with TDP-43 pathology (FTD-TDP) from FTD with tau pathology (FTD-TAU) is an avenue worth investigating. Follow-up immunohistochemical or neuroimaging studies of the affected brain tissue to correlate with these TDP-43 plasma concentrations are also needed.

2 | METHODS

2.1 | Anti-TDP-43 antibodies and recombinant TDP-43

Anti-human TDP-43 antibodies were acquired from Proteintech (10782-2-AP and 12892-1-AP, detecting N- and C-termini, respectively). Aliquots from both antibodies were diluted to 100 µg/mL in phosphate-buffered saline (PBS) and biotinylated for 2 hours at room temperature. Both antibodies were then dialyzed using a Slide-A-Lyzer Dialysis cassette for 2 hours at room temperature and left overnight with fresh PBS at 4°C. Recombinant TDP-43 (rTDP-43) was acquired from Abcam and stored at −80°C until needed.

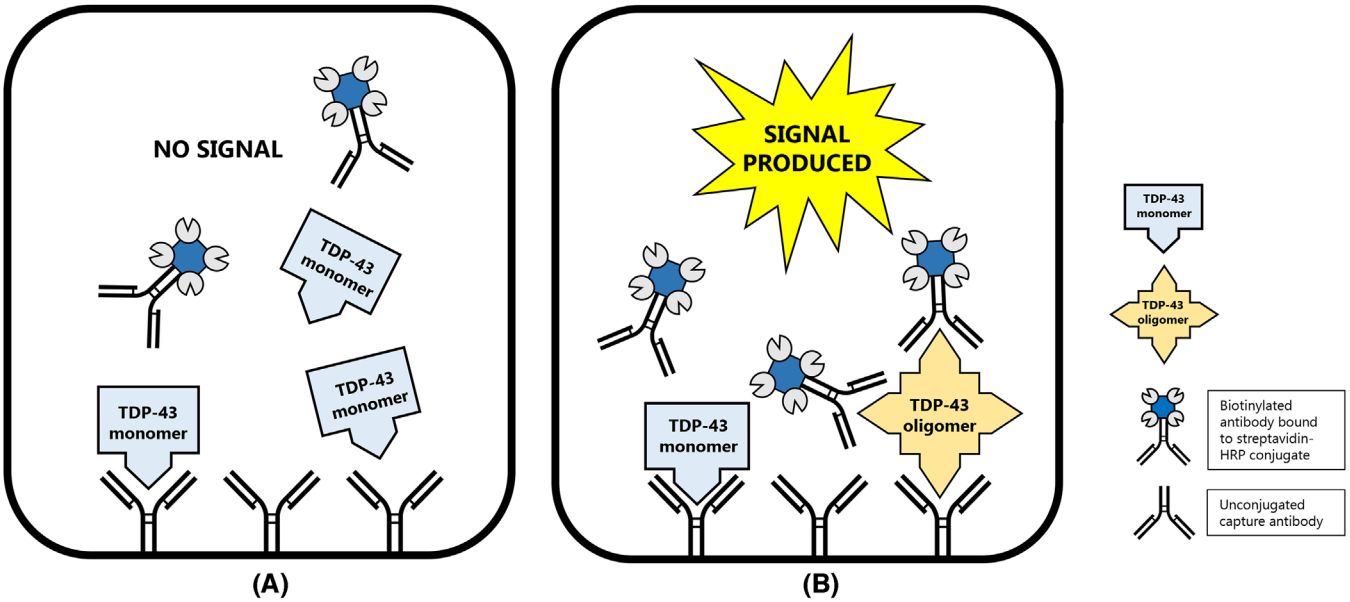

FIGURE 1 An overview of the multimer detection system. (A) The captured protein monomer has only a single epitope that is hindered by the capturing antibody. This prevents the biotinylated detection antibodies from binding to the protein and no signal is produced. On the other hand, (B) protein oligomers have multiple epitopes that remain exposed when the protein is immobilized. Biotinylated detection antibodies are free to bind to the epitope, and a signal is produced.

2.2 | Participants and sampling

Participants, including SD (n = 16), MCI (n = 18), EOAD (n = 32), LOAD (n = 16), and PD (n = 12) patients, were enrolled in the Alzheimer's Disease All Markers (ADAM) study from Seoul National University Bundang Hospital.[13] Participant demographics are summarized in Table 1. The clinical diagnosis of AD was based on the following criteria: (1) probable AD criteria proposed by the NIA-AA (National Institute on Aging–Alzheimer's Association workgroups, 2011); (2) male or female patients ages 50 to 80, (3) an educational background of at least 6 years; and (4) follow-up of at least 6 months to determine the clinical course of AD by licensed neurologists. AD was classified as either EOAD or LOAD by dividing both groups with an age at onset of 65 years. MCI diagnosis was based on the presence of impairment in one or more cognitive domains, but with preservation of functional abilities and not meeting the NIA-AA criteria for dementia due to AD. PD diagnosis was based on ^{18}F-radiolabeled N-(3-fluoropropyl)-2β-carbomethoxy-3β-(4-iodophenyl) nortropane (FP-CIT) positron emission tomography (PET) positivity. Healthy controls (n = 79) had the following criteria: (1) from a community-based population; (2) no abnormality on the Health Screening Questionnaire; (3) absence of memory complaints; (4) scoring ≤6 on the Korean Dementia Screening Questionnaire; (5) normal general cognition (within 1 SD [standard deviation] of the age- and education-adjusted norms of the Mini-Mental Status Exam [MMSE] and a score higher than 26); (6) intact activities of daily living based on Korean-Instrumental Activities of Daily Living (K-IADL ≤0.42); (7) no depression (the short-form Geriatric Depression Scale ≤7); and (8) no history of thyroid dysfunction, vitamin B12 deficiency, or folate deficiency. Exclusion criteria comprised any cognitive impairment other than AD, stroke, and delirium.

2.3 | Plasma collection

Approximately 10 mL of whole blood was drawn from each patient using a Vacutainer holder and Vacutainer ethylenediamine tetraacetic acid (EDTA) blood collection tubes. The tubes were then centrifuged within 2 hours from the blood draw at 850 × g for 30 minutes at room temperature. Forty microliters to 120 µL of the plasma was then aliquoted into purple-capped polypropylene cryotubes (Axygen by Corning), properly labeled, and stored at −80°C until further use.

2.4 | Oligomer mimicking standard protein synthesis

Imject maleimide-activated ovalbumin (OVA) was dissolved in ddH_2O to a final concentration of 10 mg/mL. The TDP-43 peptide corresponding to the target epitope was dissolved in ddH_2O to a final concentration of 5 mg/mL. Then, 50 µL of OVA was mixed with 50 µL of the target peptide and ≈300 µL of a binding buffer (0.1 M EDTA+PBS). The resulting solution was incubated for 2 hours at room temperature with rotation. The OVA in solution was then blocked with 1 µL of 120 mg/mL cysteine dissolved in acidic PBS (pH 4.5) and incubated for 1 hour at room temperature with rotation. The solution was then transferred to a dialysis cassette, and dialysis in PBS was performed for 2 hours at room temperature and transferred to 4°C for overnight incubation in fresh PBS. The resulting oligomer mimicking standard protein (OMSP) from the attachment of the target peptides to the OVA carrier protein was then collected in a microcentrifuge tube and stored in 4°C until later use.

TABLE 1 Patient demographics.

	Disease group						
			Alzheimer's disease		Parkinson's disease	Semantic dementia	
Patient characteristics	Normal ($n = 72$)	MCI ($n = 17$)	EOAD ($n = 30$)	LOAD ($n = 16$)	($n = 12$)	SD-AD ($n = 6$)	SD-FTD ($n = 10$)
Age (y)							
40–49	5	0	1	0	0	0	0
50–59	20	2	19	0	2	0	2
60–69	35	3	10	3	2	1	4
70–79	11	9	0	9	6	4	3
80–89	1	3	0	4	2	1	1
Gender							
M	25	6	12	6	8	3	3
F	47	11	18	10	4	3	7
MMSE Score							
0–9	0	0	9	2		2	3
10–19	0	2	10	7		2	2
20–24	1	7	9	7		1	1
25–30	71	8	2	0		1	2
Ethnicity							
East Asian (Korean)							

2.5 | ELISA formats in measuring TDP-43 concentrations in plasma

One hundred microliters of 1 μg/mL of 10782-2-AP antibodies (MDS) or 12892-1-AP (total ELISA) were coated on to black Nunc Maxisorp microwells overnight at 4°C. The microwells were then washed three times with 300 μL PBS with Tween (PBST; Thermo Scientific) and blocked with 200 μL of 25% diluted BlockAce (Bio-Rad) in PBS for 1 hour at room temperature. The microwells underwent another cycle of PBST washing before adding 100 μL of standards in the form of serially diluted OMSP (50, 10, 2, and 0 ng/mL) in PBST (MDS) or recombinant TDP-43 (50, 10, 2, and 0 ng/mL) in PBST (total ELISA). For patient or control samples, 50 μL of plasma sample was added to 150 μL of PBST in the microwells and left to incubate for 1 h at 37°C. After washing away the analyte three times with PBST, 100 μL of 0.5 μg/mL of biotinylated anti-TDP-43 (10782-1-AP) antibodies in PBST with 5% BlockAce was added next and incubated for 1 hour at 37°C in both MDS and total ELISA. The microwells were then washed again and 100 μL of streptavidin was conjugated with horseradish peroxidase (streptavidin-poly-HRP; Pierce Thermo Scientific, USA) diluted 10,000 times in PBST with 5% BlockAce was added and left to incubate for 30 minutes at room temperature. The microwells were then given a final washing before adding 100 μL of SuperSignal ELISA Pico chemiluminescent substrate (Thermo Scientific), and the generated luminescence was measured immediately using PerkinElmer Victor 3 multilabel microplate reader.

2.6 | Statistical analyses

Statistical analyses were performed using SPSS 25 (SPSS Inc., Chicago, IL). A $p < 0.05$ was considered statistically significant for all tests. The normality of distribution was assessed using the Kolmogorov-Smirnov test, and skewed distributions were transformed using the Box-Cox Method. One-way analysis of variance (ANOVA) was then used to evaluate significant differences between disease groups and normal controls. Post hoc analysis (Tukey's test) was used to identify group pairings with significant differences in concentrations. Receiver-operating characteristic (ROC) curve analysis was used to evaluate the diagnostic accuracy of the ELISA for phenotypes with statistically higher parameters compared to the normal controls.

3 | RESULTS

Plasma o-TDP-43 concentrations were significantly higher in SD patients compared to other neurodegenerative disease groups and normal controls ($p < 0.05$). Plasma o-TDP-43 concentrations ranged from 7.43 to 37.45 (mean of 23.86 ± 4.24) ng/mL in SD, 6.86 to 15.09 (mean of 9.97 ± 0.79) ng/mL in PD, 0 to 12.35 (mean of 4.89 ± 0.90) ng/mL in LOAD, 0 to 18.15 (mean of 7.03 ± 0.92) ng/mL in EOAD, 0 to 18.66 (mean of 5.07 ± 1.44) ng/mL in MCI, and 0 to 11.92 (mean of 3.26 ± 0.46) ng/mL in healthy controls. A significant difference in group variances for o-TDP-43 plasma concentrations was found (ANOVA; $p < 0.05$). Post hoc analysis (Tukey's test) of o-TDP-43 concentrations

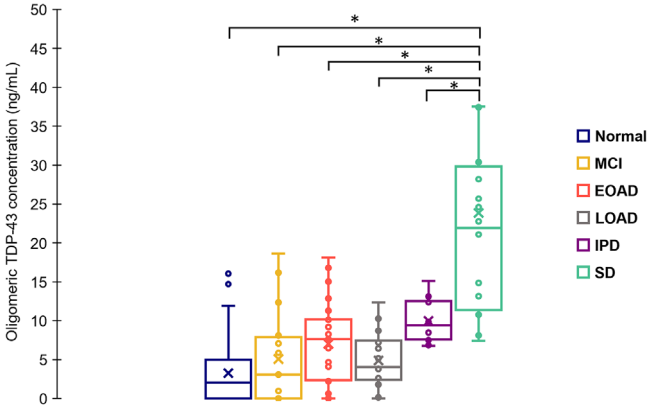

FIGURE 2 Oligomeric TDP-43 plasma concentrations in Korean patients with different neurodegenerative disorders. Tukey's test showed a statistically significant difference in oligomeric TDP-43 (o-TDP-43) plasma concentrations for patients with semantic dementia (SD) compared to all other neurodegenerative diseases including normal controls. No significant difference was found for other neurodegenerative diseases when compared to normal controls. Oligomeric TDP-43 concentrations were measured in duplicate. Disease groups are composed of normal controls, mild cognitive impairment (MCI), early-onset Alzheimer's disease (EOAD), late-onset Alzheimer's disease (LOAD), idiopathic Parkinson's disease (IPD), and semantic dementia (SD).

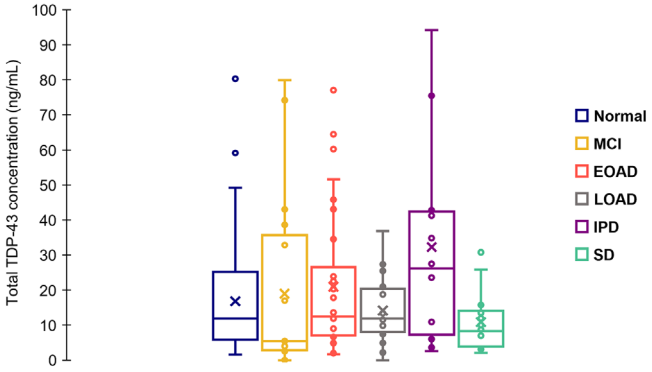

FIGURE 3 Total TDP-43 plasma concentrations in Korean patients with different neurodegenerative disorders. Analysis of variance (ANOVA) showed no significant difference in total TDP-43 (t-TDP-43) concentrations for all neurodegenerative diseases. t-TDP-43 concentrations were measured in duplicate.

showed that SD patients had significantly higher o-TDP-43 concentrations (Figure 2) compared to the other disease groups and normal controls ($p < 0.05$). No statistically significant differences were found for t-TDP-43 concentrations in all groups (Figure 3). Plotting o-TDP-43 concentrations as a ratio against the t-TDP-43 concentrations also showed a significant difference in group variances. Applying Tukey's test showed that the ratio of o-TDP-43 to t-TDP-43 was significantly higher in SD patients compared to all other groups (Figure 4).

Plasma o-TDP-43 concentrations were significantly higher in SD-FTD patients compared to other disease groups and normal controls ($p < 0.005$). Stratifying SD patients into those with comorbid FTD (SD-

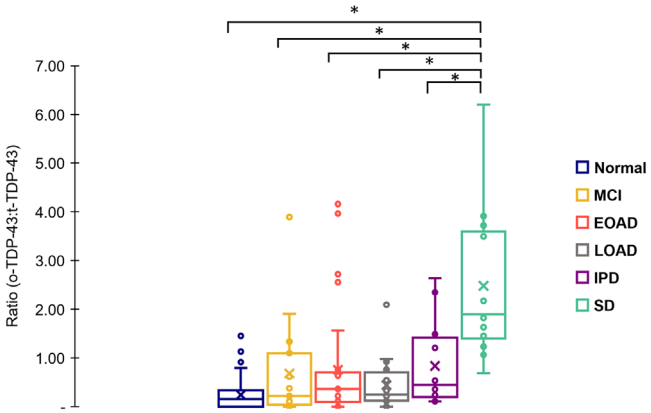

FIGURE 4 Ratios of oligomeric TDP-43 (o-TDP-43) to total TDP-43 (t-TDP-43) concentrations. Tukey's test showed a statistically significant difference in the ratio of o-TDP-43 to t-TDP-43 concentrations in semantic dementia (SD) samples compared to other neurodegenerative diseases and normal controls. No significant difference was found for other groups when compared to every other group except SD.

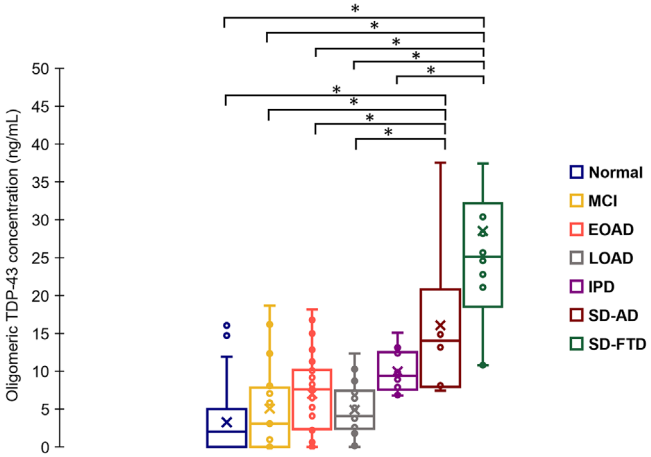

FIGURE 5 Oligomeric TDP-43 (o-TDP-43) concentrations of stratified semantic dementia (SD) samples compared to other neurodegenerative diseases. o-TDP-43 concentrations of SD-FTD (fronto-temporal dementia) samples were significantly increased compared to every other group except SD-AD (Alzheimer's disease) ($p < 0.05$). o-TDP-43 concentrations of SD-AD samples were also significantly greater than o-TDP-43 concentrations in normal controls, mild cognitive impairment (MCI), early-onset Alzheimer's disease (EOAD), and late-onset Alzheimer's disease (LOAD).

FTD) and comorbid AD (SD-AD) showed a significant difference in o-TDP-43 plasma concentrations of SD-FTD patients compared to the other disease groups (except for SD-AD) and normal controls (Figure 5). The o-TDP-43 concentrations of SD-AD patients were also significantly higher compared to the other disease groups and normal controls but were not as apparent as those o-TDP-43 concentrations from SD-FTD (Figure 5). No significant difference was observed in the o-TDP-43 plasma concentrations between SD-FTD and SD-AD patients (Figure 5). No significant differences were observed for t-TDP-43 plasma concentrations for all groups.

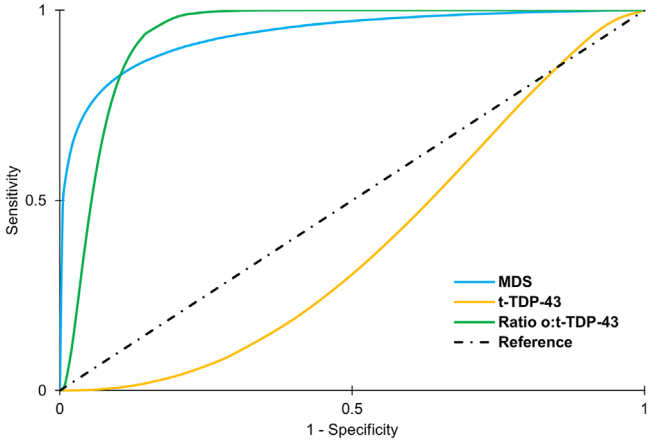

FIGURE 6 Receiver-operating characteristic (ROC) analysis of oligomeric TDP-43 (o-TDP-43) and total TDP-43 (t-TDP-43) concentrations and their ratios. Only the ratio of o:t-TDP-43 and o-TDP-43 showed excellent diagnostic performance for identifying semantic dementia (SD) samples among other groups including normal controls.

ROC analysis of o-TDP-43 showed good diagnostic accuracy for diagnosing SD among other AD pathologies and normal controls. ROC analysis for o-TDP-43 concentrations was performed to compare SD with all other groups (Figure 6). AUC was highest at 0.916 for the ratio of o:t-TDP-43 (sensitivity = 84%, specificity = 89%, positive predictive value [PPV] = 46%, and negative predictive value [NPV] = 98%), followed by o-TDP-43 at 0.904 at 95% confidence (sensitivity = 89%, specificity = 86%, PPV = 39%, and NPV = 99%). Total TDP-43 performed poorly as SD marker with AUC of 0.351 at 95% confidence.

4 | DISCUSSION

In this study, SD plasma samples were shown through MDS to have significantly higher o-TDP-43 concentrations compared to plasma collected from other disease groups ($p < 0.05$). However, no significant differences in o-TDP-43 concentrations were found in MCI, EOAD, LOAD, and PD plasma samples when compared to normal controls. In additionally, no significant differences in t-TDP-43 concentrations were found in plasma from all disease groups compared to normal controls using conventional ELISA. In a previous study by Foulds et al., significantly increased t-TDP-43 concentrations were found in the plasma of 46% FTD patients ($n = 16$) and 22% AD patients ($n = 23$) in their cohort ($n = 137$).[6] The ELISA employed in their study consisted of a monoclonal capture antibody that binds to the RRM-2 (residues 205-222) of TDP-43 and a polyclonal detection antibody that binds to the NTD (residues 1-19) and the RRM-2 region, but not overlapping with the capture antibody epitope (residues 194-213).[6]

In our study, the same polyclonal antibodies (10782-2-AP) used by Foulds et al. were applied as both capture and detection antibodies to measure o-TDP-43 levels in plasma. In contrast, different sets of polyclonal antibodies were used to capture and detect t-TDP-43. The capture antibodies used (12892-1-AP) were specific to the first 19 residues (272-290) of the low complexity domain (LCD) of TDP-43, whereas NTD and RRM-2-specific polyclonal antibodies (10782-2-AP) were used as detection antibodies. We have previously demonstrated the broad utility of MDS to quantify oligomers in other proteinopathies. In its earlier application, oligomeric prion protein (PrPd) was detected in the plasma of transmissible spongiform encephalopathy (TSE)-infected Syrian hamsters, which incorporated capture and detection antibodies specific for PrP residues 106-126 and 109-112, respectively.[9,14] Applying the same method to the plasma of healthy controls revealed no PrPd.[9] Later on, MDS was expanded to detect Aβ oligomers in the plasma of patients with AD.[10] Aβ oligomers in the plasma of AD patients were reported to be higher when compared to the plasma of the normal controls. The increased Aβ oligomer concentrations also correlated well with conventional AD biomarkers, namely the standard uptake value ratio of Pittsburg compound B (PiB SUVR) in positron emission tomography (PET) studies, the concentration of total Aβ_{42} in cerebrospinal fluid (CSF), as well as phosphorylated tau (p-tau), and total tau (t-tau) concentrations in CSF.[10] The sensitivity and specificity of MDS to differentiate AD patients from normal controls were also calculated to be 78.3% and 86.5%, respectively.[10] More recently, MDS was used as a pre-screening test for amyloid status in a cohort of 399 subjects.[15] MDS identified abnormal Aβ-PET with an AUC of 0.74 (95% CI, 0.67–0.81).[15] The MDS results combined with apolipoprotein E (*APOE*) ε4 and subject age improved the AUC to 0.81 (95% CI, 0.74–0.87).[15] This is the first attempt to use this technique in FTD plasma.

Postmortem analyses of pathological brain tissue of FTD patients in other studies showed that those diagnosed with semantic dementia presented only ubiquitin and TDP-43-positive inclusions without tau pathology, which may explain why o-TDP-43 is more pronounced in SD samples.[1,16–18] In contrast, the intrusion of tau pathology results either in the development of PNFA or the bvFTD.[4] PNFA is described as grammar and motor speech deficits wherein tau pathology is more pronounced over TDP-43 pathology. On the other hand, bvFTD is indicated by abnormal changes in behavior and personality due to frontal-predominant cortical degeneration where both tau and TDP-43 pathology have the same burden.[4]

Of interest when the SD patients were separated according to FTD or AD comorbidity, the o-TDP-43 concentrations in SD-FTD samples were significantly higher compared to the other disease groups ($p < 0.05$; Figure 5). o-TDP-43 concentrations in SD-AD samples were also significantly higher when compared to other disease groups and healthy controls ($p < 0.05$; Figure 5). However, no significant difference in o-TDP-43 concentrations was observed between the SD-FTD and SD-AD groups. The decreased o-TDP-43 plasma concentration among MCI and AD groups may be explained by the presence of other pathological proteins such as Aβ_{42} and tau neurofibrillary tangles (NFTs) in AD subtypes and MCI, which possibly sequestered o-TDP-43 into large fibrils that remain deposited inside neurons. A recent study on a *Caenorhabditis elegans* model demonstrated that the presence of TDP-43 specifically enhanced tau but not Aβ_{42} neurotoxicity, resulting in increased tau accumulation.[19] Another study that used tau-inducible cell models (iHEK) showed the accumulation of cytoplasmic TDP-43.[20] The principal role of tau in sequestering other proteins into the

cytoplasm may explain why free TDP-43 levels were not apparent and readily detectable in the plasma of patients with AD. In another study, 1D and 2D Western immunoblotting and quantitative mass spectrometry were used in paired CSF and serum samples, blood lymphocytes, brain urea fractions, and purified exosomes from CSF in patients with amyotrophic lateral sclerosis (ALS), FTD, and non-neurodegenerative diseases.[21] The TDP-43 CSF to blood concentration ratio was found to be 1:200, and it was concluded that TDP-43 in CSF can be secreted from blood. Therefore, measurement of TDP-43 concentrations was confined to plasma samples in this study.

Based on the exploratory calculated AUC values, the ratio of o-TDP-43 to t-TDP-43 concentrations in the plasma of SD patients performed well as a diagnostic marker to differentiate TDP-43 proteinopathy in SD-FTD patients with an AUC value of 0.916 followed by o-TDP-43 concentrations alone (AUC of 0.904). On the other hand, t-TDP-43 concentrations performed poorly as a diagnostic marker, with an AUC value of 0.351. Other confounding factors, such as a compromised blood-brain barrier (BBB), which alters actual free protein concentrations in blood, should still be considered in similar studies because this was considered by Foulds et al. when increased t-TDP-43 plasma concentrations were correlated with FTD diagnosis. Despite this, immunohistochemistry of brain tissue is still considered the gold standard for identifying TDP-43 proteinopathy.[22] However, this study was limited by an inability to do follow-up autopsy examinations of expired patients due to rigid ethical regulations.

5 | CONCLUSION

The practicality of MDS as an approach to differentiate and quantify o-TDP-43 concentrations in plasma was demonstrated in this study. Through MDS, patients with SD were shown to have significantly higher o-TDP-43 concentrations compared to other neurodegenerative diseases, and this was consistent with what was reported in previous studies. The difference in o-TDP-43 concentrations becomes more apparent in SD samples comorbid with FTD, further supporting TDP-43 as the major pathological marker of FTD when compared to AD. Still, the application of this approach to discriminate TDP-FTD from tau-FTD warrants investigation along with the influence of other comorbidities, and follow-up immunohistochemical studies of brain tissue to help establish plasma TDP-43 immunoassays as a reliable diagnostic approach for SD-FTD.

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CONFLICT OF INTEREST STATEMENT

The authors do not have any conflicts of interest to declare.

CONSENT STATEMENT

Plasma samples were collected with the informed consent of the patients or through their authorized representatives.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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