Diagnostic Utility of MOG Antibody Testing in Cerebrospinal Fluid

Vyanka Redenbaugh, MB, B.Ch, BAO [1,2] James P. Fryer, MS,[2]
Laura Cacciaguerra, MD, PhD [1] John J. Chen, MD, PhD [1,3]
Tammy M. Greenwood, MS, MHA,[3] Michael Gilligan, MB, BCh,[1,2,4]
Smathorn Thakolwiboon, MD [1] Masoud Majed, MD,[1] Nicholas H Chia, MBBS,[1]
Andrew McKeon, MD [1,2] John R. Mills, PhD,[2] A. Sebastian Lopez Chiriboga, MD [5]
Jan-Mendelt Tillema, MD,[1] Binxia Yang, PhD,[2] Yahya Abdulrahman, BS,[2] Kai Guo, PhD,[2]
Nisa Vorasoot, MD [1,2,6] Cristina Valencia Sanchez, MD, PhD [7]
Deena A. Tajfirouz, MD [1,3] Michel Toledano, MD,[1]
Anastasia Zekeridou, MD, PhD [1,2] Divyanshu Dubey, MD [1,2]
Grace Y Gombolay, MD [8] César Caparó-Zamalloa, MD,[9] Ilya Kister, MD,[10]
Sean J. Pittock, MD [1,2] and Eoin P. Flanagan, MB, B.Ch [1,2]

Objective: The aim of this study was to assess the diagnostic utility of cerebrospinal fluid (CSF) myelin oligodendrocyte glycoprotein antibodies (MOG-IgG) testing.
Methods: We retrospectively identified patients for CSF MOG-IgG testing from January 1, 1996, to May 1, 2023, at Mayo Clinic and other medical centers that sent CSF MOG-IgG for testing including: controls, 282; serum MOG-IgG positive MOG antibody-associated disease (MOGAD), 74; serum MOG-IgG negative high-risk phenotypes, 73; serum false positive MOG-IgG with alternative diagnoses, 18. A live cell-based assay assessed CSF MOG-IgG positivity (IgG-binding-index [IBI], ≥2.5) using multiple anti-human secondary antibodies and end-titers were calculated if sufficient sample volume. Correlation of CSF MOG-IgG IBI and titer was assessed.
Results: The pan-IgG Fc-specific secondary was optimal, yielding CSF MOG-IgG sensitivity of 90% and specificity of 98% (Youden's index 0.88). CSF MOG-IgG was positive in: 4/282 (1.4%) controls; 66/74 (89%) serum MOG-IgG positive MOGAD patients; and 9/73 (12%) serum MOG-IgG negative patients with high-risk phenotypes. Serum negative but CSF positive MOG-IgG accounted for 9/83 (11%) MOGAD patients, and all fulfilled 2023 MOGAD diagnostic criteria. Subgroup analysis of serum MOG-IgG low-positives revealed CSF MOG-IgG positivity more in MOGAD (13/16[81%]) than other diseases with false positive serum MOG-IgG (3/15[20%]) ($p = 0.01$). CSF MOG-IgG IBI and CSF MOG-IgG titer (both available in 29 samples) were correlated (Spearman's $r = 0.64$, $p < 0.001$).

View this article online at wileyonlinelibrary.com. DOI: 10.1002/ana.26931

Received Aug 23, 2023, and in revised form Mar 9, 2024. Accepted for publication Mar 15, 2024.

Address correspondence to Eoin P. Flanagan, Consultant in Neurology and Laboratory Medicine and Pathology, Professor of Neurology, Mayo Clinic College of Medicine, 200 1 St. SW Rochester, MN 55905. E-mail: flanagan.eoin@mayo.edu

From the [1]Department of Neurology and Center for Multiple Sclerosis and Autoimmune Neurology, Mayo Clinic College of Medicine, Rochester, Minnesota, USA; [2]Department of Laboratory Medicine and Pathology, Mayo Clinic College of Medicine, Rochester, Minnesota, USA; [3]Ophthalmology, Mayo Clinic College of Medicine, Rochester, Minnesota, USA; [4]Department of Neurology, St Vincent's University Hospital, Dublin, Ireland; [5]Department of Neurology, Mayo Clinic College of Medicine, Jacksonville, Florida, USA; [6]Division of Neurology, Department of Medicine, Faculty of Medicine, Khon Kaen University, Khon Kaen, Thailand; [7]Department of Neurology, Mayo Clinic, Scottsdale, Arizona, USA; [8]Emory University, Children's Healthcare of Atlanta: Pediatrics Institute, Atlanta, Georgia, USA; [9]Basic Research Center in Dementia and Central Nervous System Demyelinating Diseases, Instituto Nacional de Ciencias Neurológicas, Lima, Peru; and [10]Department of Neurology, Comprehensive MS Center, NYU Grossman School of Medicine, New York, New York, USA

Additional supporting information can be found in the online version of this article.

Interpretation: CSF MOG-IgG testing has diagnostic utility in patients with a suspicious phenotype but negative serum MOG-IgG, and those with low positive serum MOG-IgG results and diagnostic uncertainty. These findings support a role for CSF MOG-IgG testing in the appropriate clinical setting.

ANN NEUROL 2024;96:34–45

Introduction

Myelin oligodendrocyte glycoprotein antibody (MOG-IgG) was identified as a biomarker of central nervous system (CNS) demyelinating disease in 2007.[1] More recently, MOG-IgG-associated disease (MOGAD) was recognized as a separate disease, distinct from multiple sclerosis (MS) and aquaporin-4-IgG positive neuromyelitis optica spectrum disorder (AQP4 + NMOSD) with international diagnostic criteria proposed in 2023.[2] MOGAD is commonly associated with optic neuritis (ON), acute disseminated encephalomyelitis (ADEM), myelitis, cerebral monofocal or polyfocal deficits, brainstem or cerebellar deficits, cerebral cortical encephalitis, or combinations thereof.[2–8]

Cell-based assays are optimal for detection of MOG-IgG in serum, and live cell-based assays have advantages over fixed assays.[9,10] MOG-IgG cell-based assays in serum have high specificity (≈98%) for MOGAD-compatible clinical syndromes but false-positives occur more commonly than with AQP4-IgG, particularly with low-positive results or when ordered in situations with low pre-test probability.[11,12] Reducing false-positive results is important to prevent harm from misdiagnosis and side effects of unnecessary treatment. To date, MOG-IgG testing has primarily been in serum. However, recent studies suggest that MOG-IgG in CSF may have diagnostic and prognostic utility.[13–17] Thus, CSF MOG-IgG testing was incorporated into the 2023 MOGAD diagnostic criteria although with additional supporting features required.[2] Studies assessing if patients with serum negative and CSF positive MOG-IgG fulfill diagnostic criteria are lacking. Data from the US population are limited to a small subset of a prior case series.[17] CSF MOG-IgG has been identified occasionally in patients with MS and other neurologic diseases, but there are limited data on its frequency among inflammatory and non-inflammatory neurologic disease controls.[17,18] Herein, we sought to determine its diagnostic utility by assessing MOG-IgG in CSF in a large cohort of patients with inflammatory demyelinating diseases of the CNS and controls.

Methods

Standard Protocol Approvals, Registrations, and Patient Consents

The study was approved by the Mayo Clinic Institutional Review Board (IRB number 08–007846). All patients or their guardians provided consent for the use of their clinical records and biological specimens for research purposes through the Mayo Clinic Centre for Multiple Sclerosis and Autoimmune Neurology (CMSAN) biorepository or at the respective medical facility.

Identification of Inflammatory and Non-inflammatory Control Samples

CSF samples from Mayo Clinic patients available in the Neuroimmunology Laboratory were used as disease controls. A total of 282 controls were identified of which 117 had paired serum available and were negative for MOG-IgG. The remaining (n = 165) did not have paired serum available as their syndrome was not compatible with MOGAD (e.g., normal pressure hydrocephalus [NPH]). Of 282 controls, 161 were inflammatory (MS [n = 82], AQP4 + NMOSD [n = 42], neurosarcoidosis [n = 17], other antibody-mediated encephalitis [n = 6], infection [n = 5], cerebral vasculitis [n = 3], neuro-Behçet [n = 2], paraneoplastic neurological syndrome [n = 2], chronic lymphocytic inflammation with pontine perivascular enhancement responsive to steroids [CLIPPERS, n = 2]), and 121 were non-inflammatory controls (NPH [n = 71], CNS neoplasms [n = 12], idiopathic intracranial hypertension [n = 8], migraine [n = 7], cervical spondylosis [n = 5], non-inflammatory peripheral neuropathy [n = 5], degenerative neurological disease [n = 5], ischemic stroke [n = 4], epilepsy [n = 2], and spinal dural arteriovenous fistula [n = 2]).

Identification of MOGAD Patients with Serum MOG-IgG Positivity

Mayo Clinic patients from January 1, 1996, to May 1, 2023, with positive MOG-IgG in serum and MOGAD diagnosis,[2] were identified from our neuroimmunology database. CSF MOG-IgG was assessed in those with sufficient CSF volumes available (n = 74). Of these patients, 26 have been previously reported.[17]

Identification of High-Risk Phenotypes Negative for Serum MOG-IgG

Mayo Clinic databases were searched to retrospectively identify patients from January 1, 1996, to July 1, 2021 with a high-risk syndrome that tested negative for serum MOG-IgG and AQP4-IgG, and with sufficient CSF available. Additionally, prospective patients were included from July 1, 2021, to May 1, 2023, from Mayo Clinic, Emory University, New York University Langone Health and Instituto Nacional de Ciencias Neurológicas, Perú.

Mayo Clinic databases were searched to identify consecutive patients with the following clinical syndromes which were deemed as high-risk: recurrent ON (n = 42)[19]; ADEM or ADEM-like syndrome (n = 19)[6]; longitudinally-extensive myelitis (n = 11)[20]; or seronegative neuromyelitis optica spectrum disorder (NMOSD, n = 1).[21]

Identification of Those with Serum MOG-IgG Positivity but an Alternative Diagnosis

Mayo Clinic patients from January 1, 1996, to May 1, 2023, with positive MOG-IgG in serum (n = 18) who did not have MOGAD and had an alternative final diagnosis were identified and included: MS (n = 6), spinal cord infarct (n = 2), amyotrophic lateral sclerosis (n = 2), genetic/mitochondrial disorder (n = 2), non-inflammatory peripheral neuropathy (n = 2), or other phenotypes not resembling MOGAD (n = 4).

Testing Methodology

CSF assay methodology was similar to serum MOG-IgG assay methodology as described previously.[12] An in-house flow cytometry assay was used for quantitative detection of MOG-IgG on a mixed population of transfected and nontransfected cells. Heat-inactivated patient CSF (at 56°C for 35 minutes) was added to live human embryonic kidney (HEK) 293 cells transiently transfected with full-length recombinant human MOG (coexpressing nonlinked green fluorescent protein [GFP]).[6] CSF was tested at 1:2 dilution in duplicate for each sample, using 3 different anti-human-IgG secondary antibodies (Data S1). The median fluorescence intensity was determined for both nontransfected and transfected cells. The ratio of median fluorescence intensity values for GFP positive and GFP negative cells is the IgG binding index (IBI). An IBI score of 2.5 or greater (same cutoff for serum) was considered positive (Fig 1). Alternative cutoffs for positivity were analyzed, but 2.5 had optimal sensitivity and specificity (data not shown). In those with sufficient sample available, CSF was titrated in doubling dilutions. The farthest dilution yielding a positive result (IBI ≥2.5) was recorded as the endpoint of positivity. We also assessed for white matter staining on a mouse brain tissue composite using an immunofluorescence assay (Data S1).

Details of Samples

Only 1 sample per patient was analyzed. For all patients tested for serum MOG-IgG, time-matched paired CSF samples (within 1 month of each other) were used (Fig 2). The timing from clinical attacks to serum and CSF sampling in MOGAD patients was also calculated in in those with details of exact timing from onset available (Fig 2). Data on treatments utilized around the time of sampling were also abstracted and recorded if available.

Clinical Characteristics

Demographic and clinical data were abstracted from the electronic medical records by a neurologist blinded to serum and CSF MOG-IgG results. Relapses were defined as neurological disturbances of more than 24 hours duration occurring after a period of clinical remission of more than 1 month. The Expanded Disability Status Scale (EDSS) score at last follow-up was retrospectively calculated from medical record review.

Statistical Analysis

Continuous and categorical variables were reported as median (range) and number (%), respectively. Pairwise comparisons were performed using the Wilcoxon rank sum test for continuous variables and Fisher's exact test for categorical variables. A p-value <0.05 was considered statistically significant (BlueSky statistics 7.4). Spearman's rank correlation was used for variables that were not normally distributed. For sensitivity calculations, the denominator for total MOGAD patients was 83 (serum MOG-IgG positive MOGAD [n = 74] + serum MOG-IgG negative CSF MOG-IgG positive MOGAD [n = 9]). For specificity calculations, the denominator for total negatives was 363 (controls [n = 282]; high-risk phenotypes negative in serum and CSF, [n = 64]; false positive MOG-IgG in serum [n = 18]). Youden's Index (YI) was calculated as (sensitivity + sensitivity) − 1.

Results

The demographics and results for the 4 groups tested for MOG-IgG in CSF are provided in Table 1.

MOG-IgG in CSF Using the pan-IgG Fc Specific Secondary Antibody Had High Sensitivity and Specificity

We found the pan-IgG Fc-specific anti-human secondary antibody was optimal and details of these analyses and the rarely visible mouse tissue white matter immunostaining are outlined in the Data S1 and Figures S1 and S2. The pan-IgG Fc-specific anti-human secondary antibody was utilized for all the remaining analyses in this study. CSF MOG-IgG positivity (IBI ≥2.5) had a sensitivity of 90% (75/83) and specificity was 98% (356/363) with a YI of 0.88.

MOG-IgG Was Rarely Detected among Controls

There were 4/282 (1.4%) control CSF samples that were positive (median IBI 6.7, range 5.48–10.62) for MOG-IgG (Fig 3 and Table S1). Diagnoses in false-positive CSF

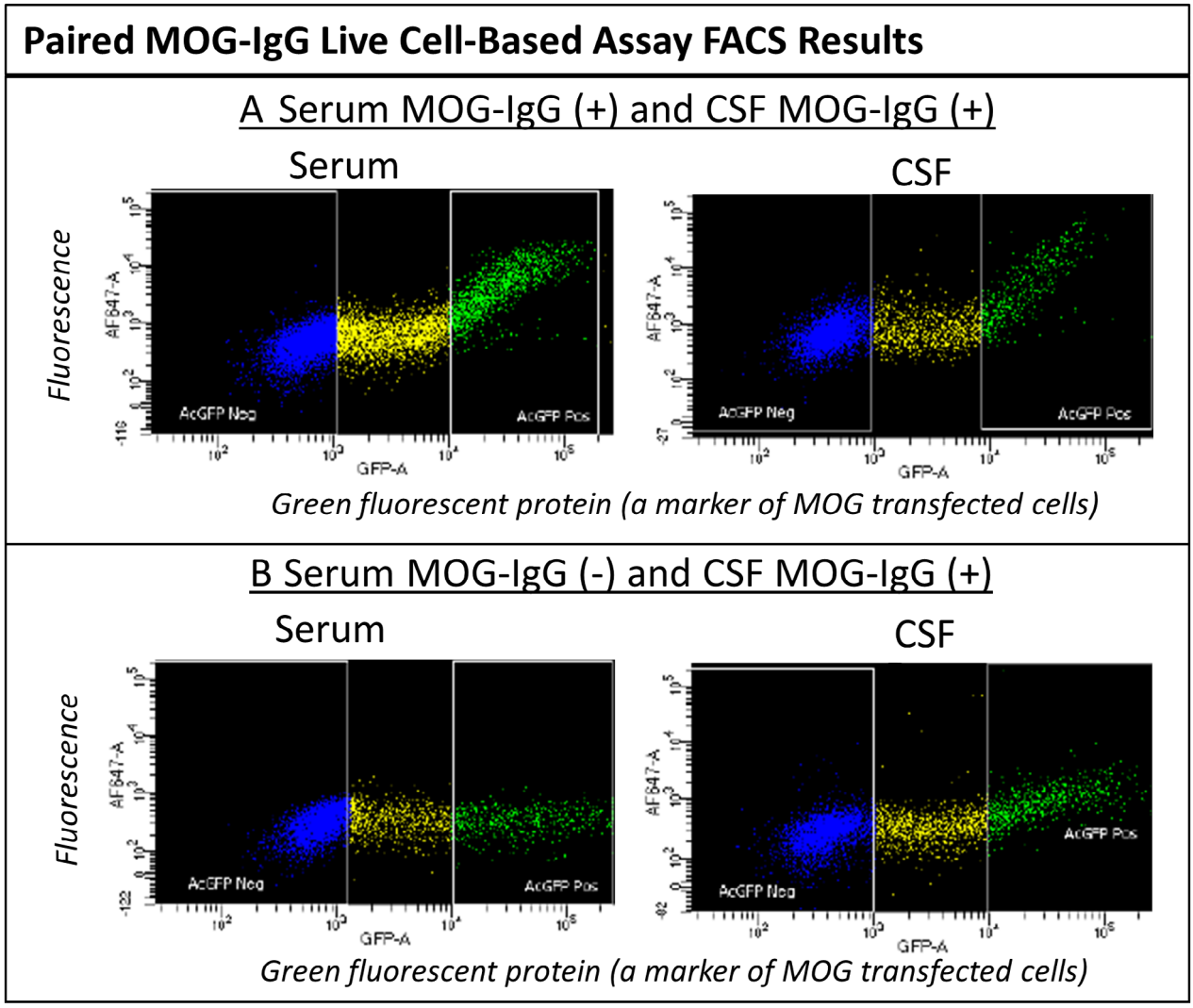

FIGURE 1: Plots of selected serum and cerebrospinal fluid MOG-IgG live cell-based assay results using fluorescence-activated cell sorting. Examples of fluorescence-activated cell sorting (FACS) live cell-based assay results of MOG-IgG in the following: (A) MOGAD patient MOG-IgG positive (immunoglobulin binding index [IBI] ≥ 2.5) in serum and CSF; (B) MOGAD patient MOG-IgG negative (IBI <2.5) in serum but positive (IBI ≥2.5) in CSF (B). Non-transfected cells are in blue and MOG-transfected cells are in green. AF647 = alexa flour 647 dye; CSF = cerebrospinal fluid; FACS = fluorescence activated cell-sorting; GFP, green fluorescent protein; IBI = immunoglobulin binding index; MOG = myelin oligodendrocyte glycoprotein.

MOG-IgG among controls were: MS, adenocarcinoma with intracranial metastases, generalized epilepsy, and spinal cord infarct. All 4 patients had negative serum MOG-IgG.

Most MOGAD Patients with Positive Serum MOG-IgG Were Also Positive in CSF

There were 74 MOGAD patients with serum MOG-IgG positivity (Table 3), with a median serum IBI of 23.44 (range 2.57–260.3) and median serum titer of 1:100 (range 1:20–1:10,000). Of these, 66 (89%) were also positive for MOG-IgG in CSF with median CSF IBI 11.18 (range 2.6–80.4), and among 26 with available CSF titers the median was 1:16 (range 1:2–1:2048) (Fig 5). An example of the accompanying MRI features of a patient with dual serum and CSF MOG-IgG positivity is shown in Fig 4.

CSF MOG-IgG Positivity Was Identified in High-Risk Phenotypes with Serum MOG-IgG Negativity, and All Fulfilled Diagnostic Criteria for MOGAD

We identified CSF MOG-IgG positivity in 9 of 73 (12%) patients (Table 2) with high-risk phenotypes but serum MOG-IgG negativity. In those with serum MOG-IgG negative and CSF MOG-IgG positive, all fulfilled

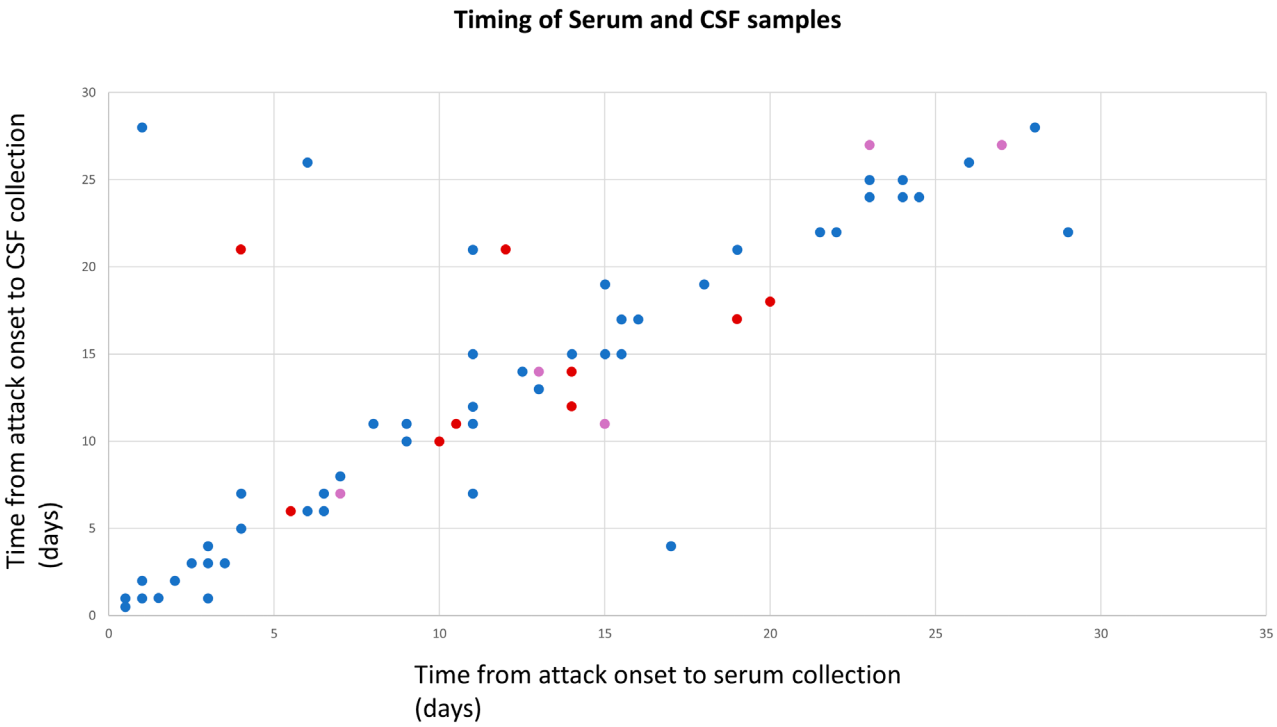

FIGURE 2: Timing of serum and CSF sample collection in MOGAD patients in 62 patients with data available. Each colored dot represents a single patient with the following color code: pink = serum MOG-IgG (+) and CSF MOG-IgG (−); blue = serum MOG-IgG (+) and CSF MOG-IgG (+); red = serum MOG-IgG (−) and CSF MOG-IgG (+). (−) = negative; (+) = positive; CSF = cerebrospinal fluid; MOG-IgG = myelin oligodendrocyte glycoprotein antibody.

TABLE 1. Characteristics of the 4 Groups of Patients Tested for CSF MOG-IgG

Parameter	Controls (n = 282)	Serum MOG-IgG Positive MOGAD (n = 74)	Serum MOG-IgG Negative, Clinically Suspicious for MOGAD (n = 73)	Serum MOG IgG False Positives (n = 18)
Median age (range)	53 (2–87)	33 (3–71)	45 (4–78)	52 (14–69)
Female sex	170 (60%)	41 (55%)	40 (55%)	11 (61%)
CSF MOG-IgG positive	4 (1.4%)	66 (89%)	9 (12%)	4 (22%)

Abbreviations: CSF = cerebrospinal fluid; EDSS = Expanded Disability Status Scale; MOG = myelin oligodendrocyte glycoprotein; MOGAD = myelin oligodendrocyte glycoprotein antibody-associated disease.

diagnostic criteria for MOGAD by having AQP4-IgG negativity and supportive clinical or MRI features as outlined in Table 2. Serum MOG-IgG negative and CSF MOG-IgG positive accounted for 9/83 (11%) MOGAD patients. Clinical features and serology details (IBI and titers) from these patients are summarized in Table 2 and TABLE 3, and radiologic features are illustrated in Fig 4. While more patients with CSF MOG-IgG had a relapsing course ($p = 0.06$), there were no statistically significant differences between patients positive for MOG-IgG in CSF only, serum only, or both serum and CSF (Table 3). Titration of serum MOG-IgG in 6 cases with serum MOG-IgG negative CSF MOG-IgG positive excluded the prozone effect as an explanation for the negative result in serum.[22,23]

Timing of Samples in Patients with MOGAD

The time between serum and CSF MOG-IgG samples is outlined in Figure 2. There was no difference in the time from attack onset to serum and CSF MOG-IgG testing or the frequency of use of acute treatment across the subgroups of MOGAD patients (Table 3). The timing

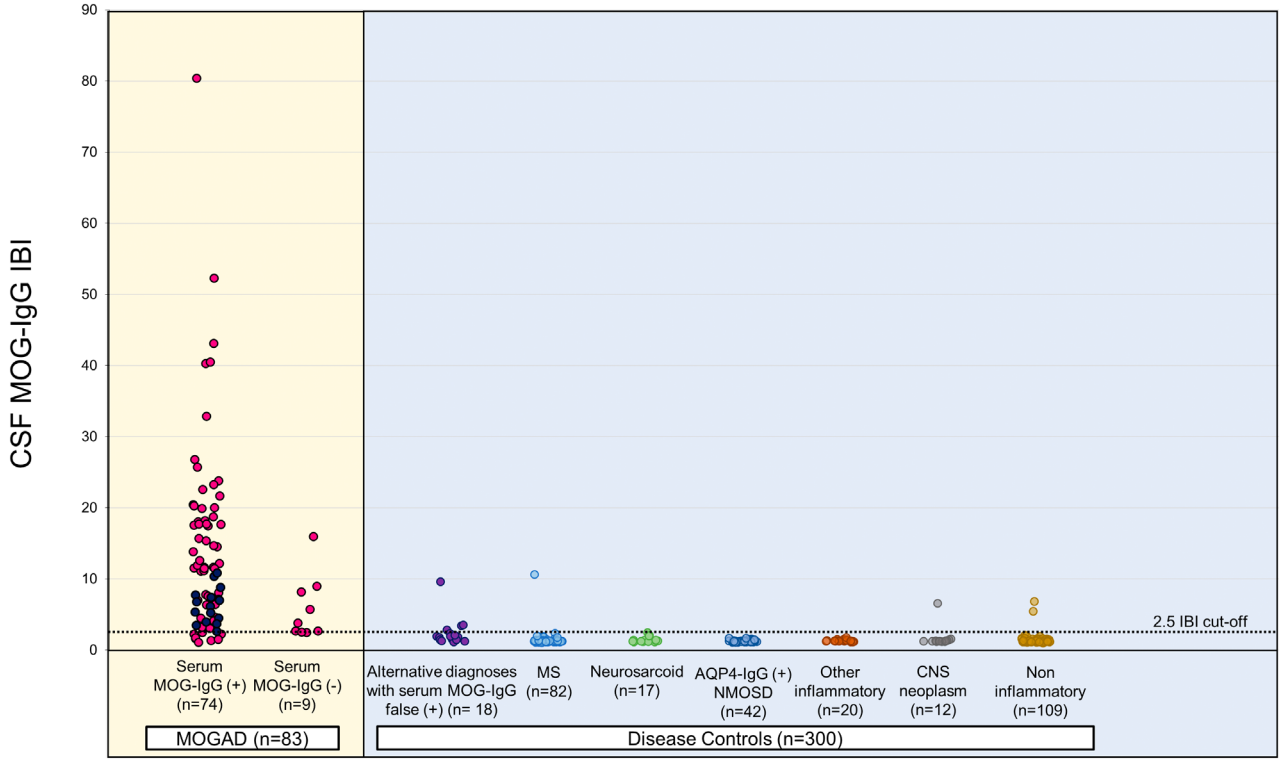

FIGURE 3: CSF MOG IgG-Binding-Index (IBI) of different samples tested. This figure illustrates the CSF IBI results (y axis) from MOGAD patients and controls (x axis) with the cutoff of an IBI of 2.5 illustrated by the dotted line. The first group (yellow shading) includes patients with MOGAD with MOG-IgG positivity in serum alone, serum and CSF, or CSF alone. The second group (light blue shading) includes patients with alternative diagnoses that had false-positive serum MOG-IgG, negative serum MOG-IgG or were not tested due to low clinical suspicion (eg, normal pressure hydrocephalus). Each colored dot reflects the CSF MOG-IgG result from each patient. Serum MOG-IgG (+) MOGAD patients with a serum titer of 1:20 or 1:40 that required supporting criteria for MOGAD diagnosis are highlighted by Navy dots. (–) = negative; (+) = positive; AQP4-IgG = aquaporin-4 antibody; CNS = central nervous system; CSF = cerebrospinal fluid; IBI = immunoglobulin-G binding index; MOGAD = myelin oligodendrocyte glycoprotein antibody-associated disease; MOG-IgG = myelin oligodendrocyte glycoprotein antibody; MS = multiple sclerosis.

also did not influence the CSF MOG-IgG IBI (data not shown).

Most False-Positive MOG-IgG in Serum Had Negative CSF MOG-IgG

There were 18 patients positive for serum MOG-IgG-without MOGAD clinico-radiologic phenotype who were found to have an alternative diagnosis (false positive). They had a median IBI in serum of 3.17 (range, 2.5–13.43) and median titer 1:40 (range, 1:20–1:100). Only 4/18 (22%) were also positive for MOG-IgG in CSF (median IBI in CSF positives of 3.51 [range, 2.83–9.62]). In a subgroup analysis of those with low-positive serum MOG-IgG (titer <1:100, IBI <10) included in this study, CSF MOG-IgG was detected more often in true MOGAD than other diseases with false-positive serum MOG-IgG (13/16 [81%] vs 3/15 [20%]; $p = 0.01$) (Fig S3). An MRI example from a multiple sclerosis patient with a MOG-IgG serum false positive result and CSF MOG-IgG negative results is shown in Figure 4.

CSF MOG-IgG IBI and CSF MOG-IgG Titer Were Correlated

There was a positive correlation between CSF IBI and titer in the 29 with sufficient sample available to determine end titers, Spearman's r = 0.64, $p < 0.001$ (Fig 5).

Discussion

In this study, CSF MOG-IgG was sensitive (90%) and highly specific (98%) for MOGAD diagnosis and CSF MOG-IgG was rarely identified among the large cohort of disease controls. Serum negative and CSF positive MOG-IgG occurred in 11% of MOGAD patients, supporting its diagnostic utility when there is a suspicious phenotype, but serum MOG-IgG is negative. This finding in an American population is consistent with studies from other regions.[14,17,24,25] Our study has many novel aspects including applications to the 2023 MOGAD diagnostic criteria,[2] utility of CSF MOG-IgG in those with low positive serum MOG-IgG and assessment of a variety of different anti-human secondary antibodies. The large

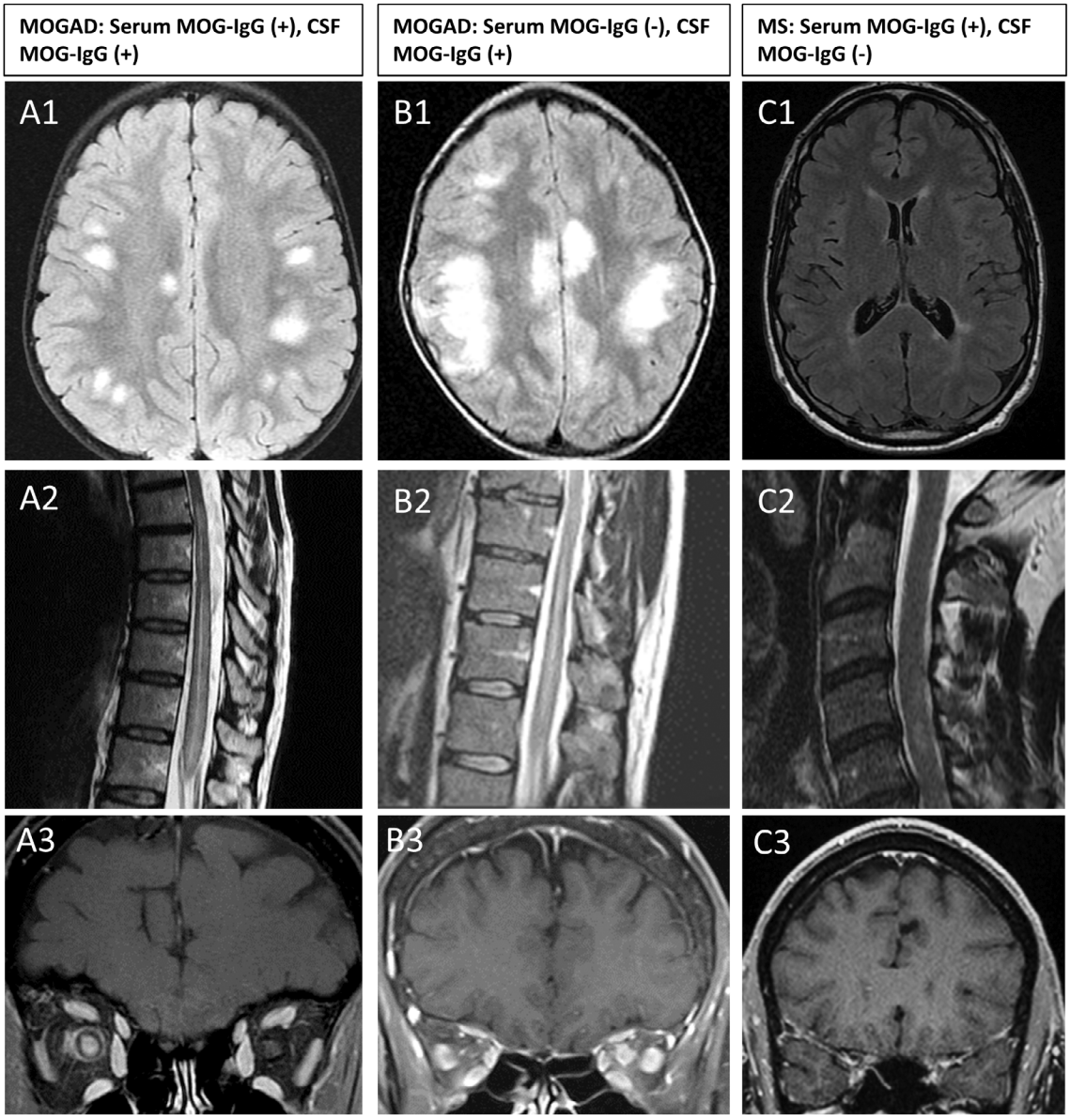

FIGURE 4: MRI examples from patients with serum or CSF MOG-IgG positivity who have either MOGAD or an alternative disease. The MRI examples shown are from: (A) MOGAD patient with serum and CSF MOG-IgG positivity; (B) MOGAD patient with serum MOG-IgG negative and CSF MOG-IgG positive; and (C) multiple sclerosis (MS) patient with MOG-IgG serum positivity but CSF MOG-IgG negative. Brain, spine, and orbital MRIs reveal ill-defined multifocal T2-hyperintensities on FLAIR images (A1, B1), longitudinally extensive T2-lesions in the thoracic spine extending to the conus (A2, B2), and anterior optic nerve enhancement (A3 [right], B3 [left]) with sheath and fatty tissue involved (A3) in MOGAD. Brain and cervical spine MRI reveal small well demarcated periventricular and splenium T2-hyperintense lesions (C1) and a short T2-lesion at the C4-C5 cervical spinal cord vertebral level (C2) and short proximal right optic nerve enhancement (C3) in MS. CSF = cerebrospinal fluid; MOGAD = myelin oligodendrocyte glycoprotein antibody-associated disease; MOG-IgG = myelin oligodendrocyte glycoprotein antibody; MS = multiple sclerosis.

number of disease controls assessed for CSF MOG-IgG is also a major strength of our study.

Serum negative and CSF positive MOG-IgG varied from 3% to 29% in previous studies,[14–17,24–26] consistent with the rate of 11% in this cohort. The 90% sensitivity of CSF MOG-IgG was slightly higher than previously reported (41–87%).[14–17,24–26] Some of these differences may reflect variability in serum and CSF MOG-IgG cut-offs, alternative anti-human secondary antibodies in serum and CSF or changes across populations studied including the proportion of adults versus children as serum negative CSF MOG-IgG positive appears to be less frequent in children.[17] It also warrants emphasis that dual MOG-IgG positivity in serum and CSF increased the likelihood of MOGAD diagnosis, suggesting CSF MOG-IgG can help in patients with low positive serum MOG-IgG and diagnostic uncertainty.

The findings in this study have relevance for the recently published 2023 MOGAD diagnostic criteria

TABLE 2. Characteristics of MOGAD patients with only CSF MOG-IgG positivity.

Patient Number	Serum IBI[a] (1:20 dilution)	CSF IBI[a] (1:2 dilution)	CSF MOG-IgG titer	Days from serum to CSF MOG-IgG testing	Age at onset	Sex	Core clinical demyelinating event per 2023 MOGAD criteria at time of serum and CSF sampling	Any other attack(s)[b]	Supporting clinical or MRI features per 2023 MOGAD diagnostic criteria met during event at time of serum and CSF sampling	Treatment in the 4 weeks preceding testing	Relapsing disease	EDSS at last follow up	Duration of follow up in months
1	1.3	16.01	Not available	0	57	Female	ADEM	ON	Multiple ill-defined T2 hyperintense lesions in supratentorial white matter	None	Yes	2	2
2	1.85	2.73	Not available	17	11	Female	ON	ON + Myelitis; Cerebral monofocal; Myelitis	Bilateral simultaneous clinical involvement	Oral steroids	Yes	2	101
3	1.21	2.51	Not available	0	9	Female	Myelitis	Cerebral polyfocal + Myelitis; ON; ON; ON; ON	Longitudinally extensive myelitis	Oral steroids	Yes	1.5	148
4	1.07	3.85	Not available	1	46	Male	Myelitis	Myelitis	Longitudinally extensive myelitis	None	Yes	3.5	4
5	1.81	8.2	Not available	0	40	Male	Myelitis	ON + Myelitis	Longitudinally extensive myelitis	None	Yes	3.5	8
6	2.0	2.73	1:4	0	39	Male	Myelitis	Nil	Longitudinally extensive myelitis	None	No	2	6
7	1.28	9.0	1:4	0	19	Male	Cerebral monofocal or polyfocal deficits	Nil	Multiple ill-defined T2 hyperintense lesions in supratentorial white matter	IV steroids	No	0	6
8	1.1	5.74	1:2	0	29	Female	ON	Nil	Bilateral simultaneous clinical involvement	IV steroids	No	2	5
9	1.24	2.53	Not available	0	24	Female	Cerebral monofocal or polyfocal deficits	ON	Multiple ill-defined T2 hyperintense lesions in supratentorial white matter	IV steroids	Yes	1	14

Abbreviations: ADEM = acute disseminated encephalomyelitis; CSF = cerebrospinal fluid; EDSS = Expanded Disability Status Scale; IV, intravenous; IBI = immunoglobulin binding index; MOGAD = myelin oligodendrocyte glycoprotein antibody-associated disease; MOG-IgG = myelin oligodendrocyte glycoprotein antibody; ON = optic neuritis.

[a](Normal, <2.5).

[b]Individual attacks separated by semicolon.

TABLE 3. Differences in demographics, clinical aspects, treatment and outcome between patients with MOGAD and high-risk syndromes stratified by serum and CSF MOG-IgG results.

	Serum MOG-IgG (+) CSF MOG-IgG (-) (n = 8)	Serum MOG-IgG (+) CSF MOG-IgG (+) (n = 66)	Serum MOG-IgG (-) CSF MOG-IgG (+) (n=9)	Serum MOG-IgG (-) CSF MOG-IgG (-) (n = 64)	P value for CSF MOG-IgG (+) vs CSF MOG-IgG (-)	P value for Serum MOG-IgG (-) CSF MOG-IgG (+) vs Serum MOG-IgG (-) CSF MOG-IgG (-)
Demographics						
Median age at CSF testing (range)	34 (17-66)	33 (3-71)	29 (9-57)	46 (4-78)	0.43	0.25
Female sex (%)	4 (50%)	37 (56%)	5 (56%)	35 (55%)	1	1
Children (%)	1 (13%)	12 (18%)	2 (22%)	7 (11%)	1	0.31
Clinical phenotypes[a]						
Optic neuritis	4 (50%)	31 (47 %)	2 (22%)	37 (58%)	1	0.07
Transverse myelitis	3 (38%)	14 (21%)	4 (44%)	18 (28%)	0.4	0.43
Encephalitis	1 (13%)	9 (14%)	3 (33%)	11 (17%)	1	0.36
Brainstem/cerebellar syndrome	0 (0%)	4 (6%)	0 (0%)	4 (6%)	1	1
Additional details						
Median age of disease onset (range)	48 (34-66)[b]	38 (3-71)[c]	29 (9-57)	42 (4-77)[d]	0.36	0.22
Days from attack onset to CSF collection	13 (6-27)[b]	10 (1-28)[c]	22 (2-28)	24 (3-30)[d]	0.2	0.21
Days from attack onset to serum collection	12 (5-28)[b]	8 (1-29)[c]	21 (3-28)	23 (2-29)[d]	0.2	0.24
Acute immunotherapy[e] within the 4 weeks prior to sampling	2 (40%)[b]	11 (20%)[c]	5 (56%)	14 (30%)[d]	0.59	0.25
Maintenance immunotherapy at the time of sampling	0 (0%)[b]	11 (20%)[c]	0 (0%)	14 (30%)[d]	0.58	0.09
CSF OCB positive (≥2 unique bands)	0 (0%)[b]	4 (7%)[c]	1/6 (17%)	4 (9%)[d]	1	1
At last follow up						
Follow up duration (months)	23 (11-57)[b]	27 (1-374)[c]	6 (2-148)	37 (1-304)[d]	0.86	0.18
Relapsing disease	0 (0%)[b]	26 (46%)[c]	6 (67%)	32 (70%)[d]	0.06	1
Permanent blindness in at least one eye	0 (0%)[b]	3 (5%)[c]	0 (0%)	5 (11%)[d]	1	0.58
Gait aid	0 (0%)[b]	6 (11%)[c]	0 (0%)	3 (7%)[d]	1	1
Death	0 (0%)[b]	2 (4%)[c]	0 (0%)	3 (7%)[d]	1	1
EDSS median (range)	1.5 (0-4)[b]	2 (0-10)[c]	2 (0-3.5)	2 (1-10)[d]	0.71	0.37

Abbreviations: ADEM = acute disseminated encephalomyelitis; CSF = cerebrospinal fluid; EDSS = Expanded Disability Status Scale score; f/u = follow up; IBI = immunoglobulin binding index; MOG = myelin oligodendrocyte glycoprotein; MOGAD = myelin oligodendrocyte glycoprotein antibody-associated disease; MS = multiple sclerosis; NPH = normal pressure hydrocephalus; OCB = oligoclonal bands; ON = optic neuritis.

[a]Some patients had multiple attack types.

[b]Exact details were only available in 5 of 8 patients.

[c]Exact details were only available in 56 of 66 patients.

[d]Exact details were only available in 46 of 64 patients.

[e]Acute immunotherapy was steroids in all but one case (patient received IVIG). No patients with MOG IgG (+) in serum or CSF had received PLEX. Two patients in the high-risk group had received PLEX.

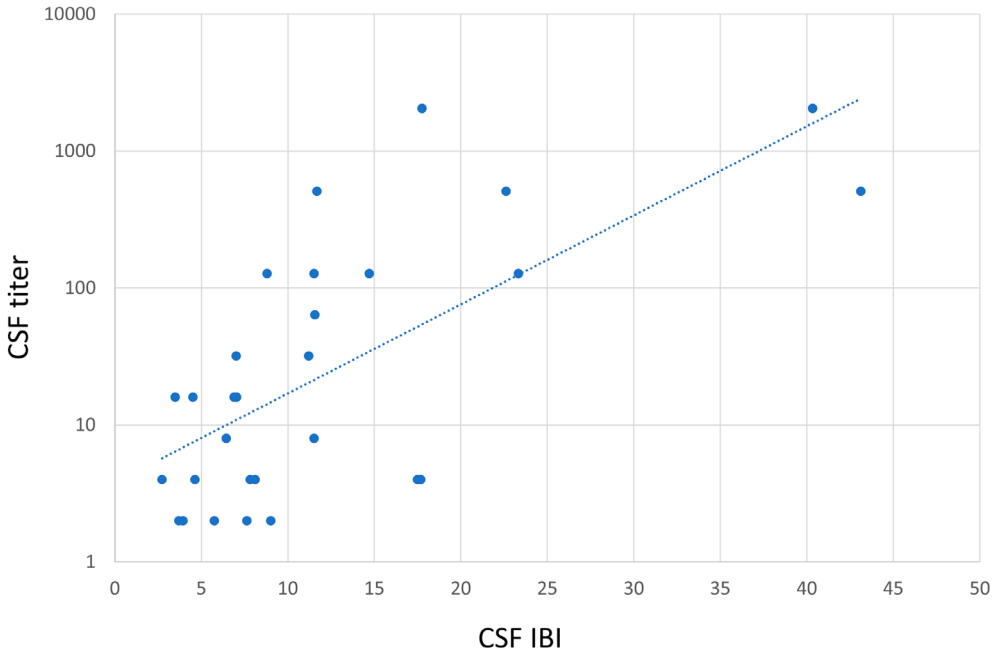

FIGURE 5: Correlation of CSF MOG-IgG IBI and titer. CSF MOG-IgG IBI and titer were correlated, spearman's r = 0.64, $p < 0.001$ in the 29 patients with sufficient sample available to assess both. CSF = cerebrospinal fluid; IBI = immunoglobulin-G binding index; MOG-IgG = myelin oligodendrocyte glycoprotein antibody.

which, in those with CSF MOG-IgG positive at any level or low positive serum MOG-IgG, require supporting clinical or MRI features that are necessary to make the diagnosis.[2] In a prior study, 1 patient with serum MOG-IgG negative and CSF MOG-IgG positive fulfilled criteria.[27] In this study, we show that all patients with a compatible phenotype and serum MOG-IgG negative and CSF MOG-IgG positive and no better diagnosis fulfilled the supportive features needed to meet MOGAD diagnostic criteria. This suggests that the 2023 MOGAD diagnostic criteria capture such patients very well.

A major strength of this study is the large number of disease controls tested, including other neurological disorders and false-positive MOG-IgG, as prior data on controls were sparse.[14,17,24,25] Our study showed CSF MOG-IgG had high specificity of 98% comparable to prior reports of 98%–100%.[14,17,25] Ultimately, there are rare patients with CSF MOG-IgG positivity without a recognized MOGAD phenotype, so the diagnosis must always consider clinical, MRI, and laboratory findings.[28] Like serum MOG-IgG, CSF MOG-IgG should only be requested in patients with high clinical suspicion for MOGAD because low positive MOG-IgG can be found in MS,[29] and since MS is much more common than MOGAD, indiscriminate testing would lead to an unacceptable number of false positives. It is notable that 4 of 9 of our serum MOG-IgG negative CSF MOG-IgG positive results just above the cutoff (IBI 2.5–3) and could be considered borderline and slight adjustments of our cutoff could have led to a lower frequency of only CSF MOG-IgG positivity. Indeed, prior assay comparison studies of serum MOG-IgG showed low positives were frequently discordant across laboratories while clear positives were highly concordant.[30] Future multicenter CSF MOG-IgG assay comparison studies will be important to determine if low positive CSF MOG-IgG results are reproducible across assays at different laboratories and for now such results require more cautious interpretation.

In a prior report, serum MOG-IgG negative and CSF MOG-IgG positive was not seen with ON and encountered predominantly with myelitis or encephalitis presentations.[25] We found most patients with serum MOG-IgG negative and CSF MOG-IgG positive had mixed phenotypes and although 2 (22%) had MOGAD ON at sampling, this latter phenotype was under-represented compared to what was expected from the high-risk syndromes screened and what we encounter with serum MOG-IgG positivity.[25] CSF-restricted oligoclonal bands were found in 17% of those with serum MOG-IgG negative and CSF MOG-IgG positive MOGAD, compared to 17–52% in prior studies.[17,25] This study may have been underpowered to prove that patients with MOG-IgG detected in CSF had more frequent relapses ($p = 0.06$) or worse clinical outcomes, as shown in other studies.[17]

This study has limitations including the retrospective nature. A live cell-based assay was used, but comparative analysis of fixed and live cell-based CSF MOG-IgG assays

are needed like has been done in serum.[9,30,31] Antibody titers are more commonly reported than IBI and the latter outnumbered titers in this study due to limited CSF volumes available; however, MOG-IgG CSF titers and CSF IBI were correlated. For paired serum and CSF analyses, up to 30 days between sample collection were allowed, which could influence our results. However, samples were generally collected early after an attack and were not influenced by plasma exchange (PLEX) which could result in discordant serum and CSF results. IVIg could potentially lead to false positive MOG-IgG serum results from antibodies present in donors or reduce signal-to-noise and lead to a serum false negative result.[32,33] While a recent analysis of MOG-IgG testing in serum after IVIg showed MOG-IgG serum antibody levels after IVIg followed the expected course, its impact on CSF MOG-IgG has not been studied.[34] While corticosteroids were more frequently used in this study prior to sampling they would be less likely to influence the results than PLEX or IVIg. Future studies should assess whether MOG-IgG persists longer in CSF than serum. Due to the limited sample volumes and differing secondary antibodies that we utilize in serum (IgG1) and CSF (pan-IgG Fc-specific), we were not able to accurately assess for intrathecal synthesis and this is an important aspect that future studies should evaluate. Finally, there was potential for selection bias as we only screened certain high-risk syndromes for CSF MOG-IgG (eg, brainstem and cerebellar deficits were not assessed) which could underestimate the benefit of CSF MOG-IgG testing. On the contrary, some of the phenotypes selected as high risk included aspects that allowed them fulfill the supporting clinical or MRI features of the 2023 MOGAD diagnostic criteria (eg, longitudinally extensive myelitis) and that could have led to over-estimation of the proportion of patients with MOG-IgG detected only in CSF that would fulfill 2023 MOGAD diagnostic criteria.

In summary, this study shows that it is useful to assess CSF MOG-IgG in clinical care when there is a phenotype suspicious for MOGAD and serum MOG-IgG is negative as it can identify approximately 10% more cases than serum testing alone. CSF MOG-IgG is also helpful in those with low positive serum MOG-IgG in whom there is diagnostic uncertainty as dual positivity in serum and CSF increases the likelihood of MOGAD.

Acknowledgement

This study was funded by an RO1 from the National Institute of Neurological Disorders and Stroke (R01NS113828). This publication was supported by CTSA Grant Number UL1 TR002377 from the National Center for Advancing Translational Science (NCATS). Its contents are solely the responsibility of the authors and do not necessarily represent the official views of the NIH. This study was supported by the Mayo Clinic Center for Multiple Sclerosis and Autoimmune Neurology.

Author Contributions

V.R., J.J.C., S.J.P., and E.P.F. contributed to the conception and design of the study; V.R., J.P.F., L.C., J.J.C., T.M.G., M.G., S.T., M.M., N.H.C., A.M., J.R.M., A.S.L.C., J.M.T., B.Y., Y.A., K.G., N.V., C.V.S., D.A.T., M.T., A.Z., D.D., G.Y.G., C.C.Z., I.K., S.J.P., and E.P.F. contributed to the acquisition and analysis of data; V.R., L.C., M.G., A.Z., S.T., and E.P.F. contributed to drafting the text or preparing the figures.

Potential Conflicts of Interest

J.R.M., A.Z., D.D., S.J.P., and E.P.F. are working in the Mayo Clinic neuroimmunology laboratory clinical service that offers MOG-IgG testing in serum and may offer MOG antibody testing in CSF in the future but none of the authors receive personal income from these tests. V.R., J.P.F., L.C., J.J.C., T.M.G., M.G., S.T., M.M., N.H.C., A.M., A.S.L.C., J.M.T., B.Y., Y.A., K.G., N.V., C.V.S., D.A.T., M.T., G.Y.G., C.C.Z., and I.K. all report no disclosures.

Data Availability

Anonymized data used for this study will be made available from the corresponding author upon reasonable request.

References

1. O'Connor KC, McLaughlin KA, De Jager PL, et al. Self-antigen tetramers discriminate between myelin autoantibodies to native or denatured protein. Nat Med 2007;13:211–217.
2. Banwell B, Bennett JL, Marignier R, et al. Diagnosis of myelin oligodendrocyte glycoprotein antibody-associated disease: international MOGAD panel proposed criteria. Lancet Neurol 2023;22:268–282.
3. Weber MS, Derfuss T, Brück W. Anti-myelin oligodendrocyte glycoprotein antibody-associated central nervous system demyelination-a novel disease entity? JAMA Neurol 2018;75:909–910.
4. de Seze J. MOG-antibody neuromyelitis optica spectrum disorder: is it a separate disease? Brain 2017;140:3072–3075.
5. Kunchok A, Chen JJ, Saadeh RS, et al. Application of 2015 seronegative Neuromyelitis Optica Spectrum disorder diagnostic criteria for patients with myelin oligodendrocyte glycoprotein IgG-associated disorders. JAMA Neurol 2020;77:1572–1575.
6. López-Chiriboga AS, Majed M, Fryer J, et al. Association of MOG-IgG Serostatus with relapse after acute disseminated encephalomyelitis and proposed diagnostic criteria for MOG-IgG-associated disorders. JAMA Neurol 2018;75:1355–1363.
7. Ramanathan S, Mohammad S, Tantsis E, et al. Clinical course, therapeutic responses and outcomes in relapsing MOG antibody-associated demyelination. J Neurol Neurosurg Psychiatry 2018;89:127–137.

8. Jurynczyk M, Messina S, Woodhall MR, et al. Clinical presentation and prognosis in MOG-antibody disease: a UK study. Brain 2017; 140:3128–3138.
9. Waters PJ, Komorowski L, Woodhall M, et al. A multicenter comparison of MOG-IgG cell-based assays. Neurology 2019;92: e1250–e1255.
10. Yeh EA, Nakashima I. Live-cell based assays are the gold standard for anti-MOG-ab testing. Neurology 2019;92:501–502.
11. Jarius S, Paul F, Aktas O, et al. MOG encephalomyelitis: international recommendations on diagnosis and antibody testing. J Neuroinflammation 2018;15:134.
12. Sechi E, Buciuc M, Pittock SJ, et al. Positive predictive value of myelin oligodendrocyte glycoprotein autoantibody testing. JAMA Neurol 2021;78:741–746.
13. Hacohen Y, Kerr W, Waters P. Intrathecal production of MOG-IgG: highlighting the need for CSF testing in clinical practice. Neurology 2021;97:12–13.
14. Kwon YN, Kim B, Kim JS, et al. Myelin oligodendrocyte glycoprotein-immunoglobulin G in the CSF: clinical implication of testing and association with disability. Neurol Neuroimmunol Neuroinflamm 2022;9:9.
15. Akaishi T, Takahashi T, Misu T, et al. Difference in the source of anti-AQP4-IgG and anti-MOG-IgG antibodies in CSF in patients with Neuromyelitis Optica Spectrum disorder. Neurology 2021;97: e1–e12.
16. Mariotto S, Gajofatto A, Batzu L, et al. Relevance of antibodies to myelin oligodendrocyte glycoprotein in CSF of seronegative cases. Neurology 2019;93:e1867–e1872.
17. Carta S, Cobo Calvo Á, Armangué T, et al. Significance of myelin oligodendrocyte glycoprotein antibodies in CSF: a retrospective multicenter study. Neurology 2023;100:e1095–e1108.
18. Armangue T, Olivé-Cirera G, Martínez-Hernandez E, et al. Associations of paediatric demyelinating and encephalitic syndromes with myelin oligodendrocyte glycoprotein antibodies: a multicentre observational study. Lancet Neurol 2020;19:234–246.
19. Jitprapaikulsan J, Chen JJ, Flanagan EP, et al. Aquaporin-4 and myelin oligodendrocyte glycoprotein autoantibody status predict outcome of recurrent optic neuritis. Ophthalmology 2018;125: 1628–1637.
20. Jitprapaikulsan J, Lopez Chiriboga AS, Flanagan EP, et al. Novel glial targets and recurrent longitudinally extensive transverse myelitis. JAMA Neurol 2018;75:892–895.
21. Jiao Y, Fryer JP, Lennon VA, et al. Updated estimate of AQP4-IgG serostatus and disability outcome in neuromyelitis optica. Neurology 2013;81:1197–1204.
22. Mathai A, Panicker S, Kannoth S, Anandakuttan A. Prozone phenomenon observed in indirect immunofluorescence assay by antibodies against neuronal antigens. J Neuroimmunol 2020;349:577415.
23. Jain D, Choudhuri J, Chauhan R, et al. False negative single antigen bead assay: is it always an effect of prozone? J Clin Lab Anal 2018; 32:e22237.
24. Pace S, Orrell M, Woodhall M, et al. Frequency of MOG-IgG in cerebrospinal fluid versus serum. J Neurol Neurosurg Psychiatry 2022;93: 334–335.
25. Matsumoto Y, Kaneko K, Takahashi T, et al. Diagnostic implications of MOG-IgG detection in sera and cerebrospinal fluids. Brain 2023; 146:3938–3948.
26. Jarius S, Ruprecht K, Kleiter I, et al. MOG-IgG in NMO and related disorders: a multicenter study of 50 patients. Part 1: frequency, syndrome specificity, influence of disease activity, long-term course, association with AQP4-IgG, and origin. J Neuroinflammation 2016; 13:279.
27. Forcadela M, Rocchi C, San Martin D, et al. Timing of MOG-IgG testing is key to 2023 MOGAD diagnostic criteria. Neurol Neuroimmunol Neuroinflamm 2024;11:e200183.
28. Sechi E, Cacciaguerra L, Chen JJ, et al. Myelin oligodendrocyte glycoprotein antibody-associated disease (MOGAD): a review of clinical and MRI features, diagnosis, and management. Front Neurol 2022; 13:885218.
29. Spadaro M, Gerdes LA, Krumbholz M, et al. Autoantibodies to MOG in a distinct subgroup of adult multiple sclerosis. Neurol Neuroimmunol Neuroinflamm 2016;3:e257.
30. Reindl M, Schanda K, Woodhall M, et al. International multicenter examination of MOG antibody assays. Neurol Neuroimmunol Neuroinflamm 2020;7:e674.
31. Cross H, Sabiq F, Ackermans N, et al. Myelin oligodendrocyte glycoprotein (MOG) antibody positive patients in a multi-ethnic Canadian cohort. Front Neurol 2020;11:525933.
32. de Beer F, Schreurs MW, Foncke EM. False positive autoantibodies to glutamic acid decarboxylase in opsoclonus-myoclonus-ataxia syndrome after intravenous treatment with immunoglobulin. Clin Neurol Neurosurg 2009;111:643–644.
33. Grüter T, Ott A, Meyer W, et al. Effects of IVIg treatment on autoantibody testing in neurological patients: marked reduction in sensitivity but reliable specificity. J Neurol 2020;267:715–720.
34. Roy S, Vasileiou E, Barreras P, et al. Longitudinal evaluation of serum MOG-IgG titers in MOGAD after initiation of maintenance immunoglobulin: a case series. Mult Scler 2023;13524585231211119.