L A P R E S S E M É D I C A L E Journal – EMR 2025 -0-0 Vol. 88 No. , Issue 2 (2025): Pages A1-A14, 101-198 (20 January 2025)

Anti-aquaporin 4 IgG (NMO–IgG) Antibodies among Sudanese Patients with Neuromyelitis Optica and Multiple Sclerosis

Marwa Alamin [1] *Malaz Alamin*[2] *Anas Ahmed* [3], *Sharif Ahmed* [4], Rayan Morad [5], *Wafaa Ali* [6]

DOI: 10.5281/zenodo.15104251

L A P R E S S E M É D I C A L E

Abstract : ***Introduction****: Neuromyelitis optica (NMO) and multiple sclerosis (MS) are distinct autoimmune demyelinating disorders of the central nervous system affecting various populations worldwide.* ***Purpose****: This study aims to assess the prevalence of NMO-IgG antibodies in Sudanese patients with NMO and compare levels to MS and other autoimmune disorders. Methods: Blood samples from 40 participants, including NMO, MS, inflammatory control groups, and 20 healthy controls, were analyzed. NMO-IgG antibodies were detected using the Euroimmune indirect immunofluorescence assay.*

Results*: NMO-IgG antibodies were present in 86.7% of the NMO group and 6.7% of the MS group and were absent in healthy and inflammatory controls, highlighting its diagnostic specificity.*

Conclusion*: These findings underscore the potential impact of integrating NMO-IgG testing into diagnostic protocols. This integration could significantly improve clinical outcomes for NMO patients in Sudan, providing a practical application of the research.*

Introduction

Neuromyelitis optica (NMO) and multiple sclerosis (MS) are autoimmune demyelinating disorders of the central nervous system (CNS) with distinct clinical and pathological characteristics. MS is primarily characterized by multifocal lesions of the CNS white matter, while NMO predominantly affects the optic nerves and spinal cord, often leading to severe disability if left untreated (Lennon et al., 2004). The discovery of a specific autoantibody targeting aquaporin-4 (AQP4), a water

Copyright reserved for the © WOS-EMR Journals - 2025

channel protein abundantly expressed in the CNS, marked a significant breakthrough in differentiating NMO from MS (Lennon et al., 2005). This antibody, termed NMO-IgG, serves as a diagnostic marker for NMO and highlights the autoimmune nature of this disease, now recognized as an AQP4-IgG seropositive spectrum disorder.

The pathophysiology of NMO involves the binding of NMO-IgG to AQP4, leading to astrocyte injury, complement activation, and subsequent demyelination and neuronal damage (Papadopoulos & Verkman, 2013). Studies indicate that early detection and treatment of NMO-IgG-positive patients are critical for preventing relapses and long-term disability (Kim et al., 2020). While extensive research has been conducted on NMO in Western and Asian populations, the prevalence, clinical characteristics, and diagnostic utility of NMO-IgG in African populations, particularly Sudanese patients, remain underexplored. This knowledge gap is significant, given the unique genetic, environmental, and healthcare system factors that may influence disease expression and outcomes in this population.

This study aims to address this gap by investigating the prevalence of NMO-IgG antibodies among Sudanese patients with NMO and comparing these findings with those from MS and other autoimmune conditions. By examining the serological landscape in Sudan, this research aims to enhance diagnostic accuracy and inform targeted treatment strategies for demyelinating central nervous system (CNS) disorders in this region, potentially improving the quality of care for these patients.

Literature Review

Neuromyelitis optica (NMO), historically known as Devic's disease, is a rare and severe autoimmune demyelinating disorder of the central nervous system (CNS), primarily characterized by the co-occurrence of optic neuritis and transverse myelitis (Wingerchuk et al., 2007; Lennon et al., 2004). Unlike multiple sclerosis (MS), which also involves central nervous system (CNS) demyelination, NMO is distinguished by its distinct pathophysiology and clinical features, underscoring the need for accurate differentiation between these conditions (Lennon et al., 2004).

A pivotal breakthrough in understanding NMO's pathogenesis was the identification of NMO-IgG, an autoantibody targeting aquaporin-4 (AQP4), a water channel protein abundantly expressed on astrocytic foot processes at the blood-brain barrier (Lennon et al., 2005; Papadopoulos & Verkman, 2013). AQP4 plays a crucial role in maintaining water homeostasis within the central nervous system (CNS), and its dysfunction leads to astrocyte damage and subsequent inflammatory demyelination (Verkman et al., 2014). The discovery of NMO-IgG has provided a reliable biomarker for NMO and its related spectrum disorders, enabling more precise differentiation from MS (Wingerchuk et al., 2015; Saiz et al., 2018).

Studies have demonstrated that NMO-IgG is detected in approximately 70-80% of patients with NMO, offering high specificity and sensitivity (Jarius & Wildemann, 2010; Cortese et al., 2022).

These antibodies are rarely found in MS, further supporting their diagnostic utility in distinguishing NMO from other Inflammatory CNS disorders (Chan, 2017; Kim et al., 2020). The specificity of NMO-IgG for NMO underscores its role as a diagnostic marker and a window into the disease's unique pathophysiology.

The clinical implications of identifying NMO as a distinct disease entity are profound. Unlike MS, treatment often includes disease-modifying therapies aimed at modulating the immune system; NMO requires targeted therapeutic approaches.

Immunosuppressive therapies, including corticosteroids, azathioprine, and rituximab, are effective in reducing relapses and improving outcomes for patients with NMO (Wingerchuk et al., 2007; Marignier et al., 2021). The recognition of AQP4-IgG's pathogenic role has also spurred the development of novel biologics targeting the complement system and AQP4 interactions, such as eculizumab and natalizumab (Pittock et al., 2019; Yamamura et al., 2019).

In summary, the identification of NMO-IgG has revolutionized the understanding, diagnosis, and management of neuromyelitis optica. It has established NMO as a distinct disease entity with specific pathogenic mechanisms and therapeutic targets, improving diagnostic precision and treatment strategies for affected individuals. Continued research into the immunopathogenesis of NMO is likely to yield further advances in personalized care and improved outcomes for patients with this challenging condition.

Methodology

This research was conducted as a cross-sectional case-control study over one year from May 2011 to May 2012. The study was divided into three phases: the first six months were dedicated to preparing laboratory materials and collecting clinical samples, the subsequent four months focused on reviewing relevant literature and documentation, and the final two months were reserved for data analysis and reporting.

The study was conducted at three clinical centres in Sudan: the Neurology Clinic at Soba University Hospital, Elshaab Teaching Hospital, and the Rheumatology Clinic at Omdurman Military Hospital. The study population consisted of patients who presented to these clinics with confirmed diagnoses of neuromyelitis optica (NMO), multiple sclerosis (MS), or other inflammatory neurological conditions, as well as a group of healthy individuals serving as the control group.

Participants were eligible for inclusion if they were between 16 and 60 years of age and had a confirmed diagnosis based on established diagnostic criteria. Individuals with NMO or MS were required to have comprehensive clinical, laboratory, cerebrospinal fluid (CSF), and magnetic resonance imaging (MRI) data available for review. Patients with specific inflammatory neurological disorders, such as systemic lupus erythematosus or Sjögren's syndrome, were also included. Those outside the defined age range or without a confirmed diagnosis or complete data were excluded from the study.

The final sample included a total of 60 participants: 15 with NMO, 15 with MS (diagnosed using the McDonald criteria), 10 with other inflammatory disorders, and 20 with healthy controls. Data collection was conducted using three structured tools: a demographic data form, a clinical symptoms questionnaire, and laboratory measurements of serum NMO-IgG antibodies.

Laboratory testing was conducted using the EUROIMMUN Indirect Immunofluorescent Test (IIFT) to detect anti-AQP4 (NMO-IgG) antibodies. This test uses BIOCHIP slides containing transfected and non-transfected cells as substrates. Serum samples were diluted at a ratio of 1:10 and incubated on the slides. After incubation, fluorescein-labelled anti-human globulin was added to detect the presence of antibodies. The results were visualized using a fluorescence microscope, and positive cases were identified based on the appearance of specific immunofluorescence patterns.

All data were entered and analyzed using the Statistical Package for the Social Sciences (SPSS), version 26. Descriptive statistics were computed for demographic and clinical variables. Chi-square tests were used to assess associations between categorical variables, and the results were presented in the form of tables and figures.

Ethical Consideration

Ethical approval for the study was obtained from the Ethical Committee of the Sudanese Medical Specialization Board (SMSB).

- Informed consent was obtained from all participants.
- Confidentiality and anonymity were maintained throughout the research process.
- All procedures were conducted by institutional ethical standards and the Declaration of Helsinki.

Result:

This study investigated the presence of anti-aquaporin-4 immunoglobulin G (NMO-IgG) antibodies among Sudanese patients with neuromyelitis optica (NMO), multiple sclerosis (MS), and other inflammatory autoimmune disorders. Blood samples were collected from 40 participants, divided into four groups: healthy controls, inflammatory controls, MS patients, and NMO patients. The mean age of participants ranged from 35.2 years in the MS group to 37.25 years in the healthy control group, with females comprising the majority across all groups, reaching 93.3% in the NMO group.

NMO-IgG antibodies were absent in all healthy controls and inflammatory controls, but they were detected in 6.7% of MS patients and 86.7% of NMO patients. These results

highlight the specificity of NMO-IgG in distinguishing NMO from MS and other inflammatory disorders. MRI findings revealed that 73.3% of NMO patients had regular brain MRIs, whereas 93.3% exhibited spinal abnormalities, predominantly longitudinally extensive transverse myelitis in the cervical spine. In comparison, 40% of MS patients demonstrated dorsal spinal changes, and only 26.7% had regular brain MRIs.

Clinical presentations varied significantly between groups. Visual impairment was universal among NMO patients, affecting 100% of the group, compared to 60% in the MS group. Walking difficulties were also more prevalent in NMO patients (93.3%) than in MS patients (73.3%). The first clinical

Presentations included lower limb weakness in 66.7% of NMO patients, whereas MS patients showed equal prevalence of lower limb weakness (33.3%) and loss of vision (33.3%). Optic neuritis occurred in 53.3% of NMO patients as a single attack and 33.3% as recurrent attacks, while in the MS group, single and recurrent attacks were observed in 20% and 26.7%, respectively.

Cerebrospinal fluid (CSF) analysis revealed elevated protein levels in over 50% of NMO patients, with no evidence of oligoclonal bands. Conversely, 96% of MS

patients exhibited oligoclonal bands, confirming their utility as a diagnostic marker for MS. Additionally, antinuclear antibodies (ANA) were detected in 13.3% of NMO patients, a lower percentage compared to global findings, potentially reflecting regional immunogenetic variations.

These findings confirm the diagnostic value of NMO-IgG antibodies in distinguishing NMO from MS and other inflammatory disorders. The results also highlight the distinct clinical and imaging characteristics of NMO in the Sudanese population, providing a basis for more targeted diagnostic and therapeutic approaches.

Table 1: Demographic and Clinical Features

Parameter	Healthy Control	Inflammatory Control	MS Group	NMO Group
Number of Participants	20	10	15	15
Mean Age (Years)	37.25	36.8	35.2	36.2
Gender (% Female)	90%	80%	73.3%	93.3%
Visual Difficulty (%)	0%	0%	60%	100%
Walking Difficulty (%)	0%	0%	73.3%	93.3%

Table 2: Diagnostic and MRI Findings

Parameter	MS Group	NMO Group
MRI Brain Normal (%)	26.7	73.3
MRI Spine Abnormal (%)	93.3 (Dorsal - 40%)	100 (Cervical - 86.7%)
Fundus Examination Normal (%)	60	0
Oligoclonal Bands in CSF (%)	60	0
AQP4 (NMO-IgG) Positive (%)	6.7	86.7
dsDNA Antibodies Positive (%)	0	0
ANA Positive (%)	6.7	13.3

Table 3: Clinical Features and Disease Onset

Parameter	MS Group	NMO Group
Disease Onset Mean Age (Years)	**30.13**	**32.67**
First Clinical Presentation - Lower Limb Weakness (%)	**33.3**	**66.7**
First Clinical Presentation - Loss of Vision (%)	**33.3**	**33.3**
Optic Neuritis - One Attack (%)	20	53.3
Optic Neuritis - Two Attacks (%)	26.7	33.3
Myelitis - One Attack (%)	33.3	53.3
Myelitis - Two Attacks (%)	33.3	26.7

MRI Findings Comparisons

Figure 1: MRI Findings Comparison (100% Stacked Bar Chart)

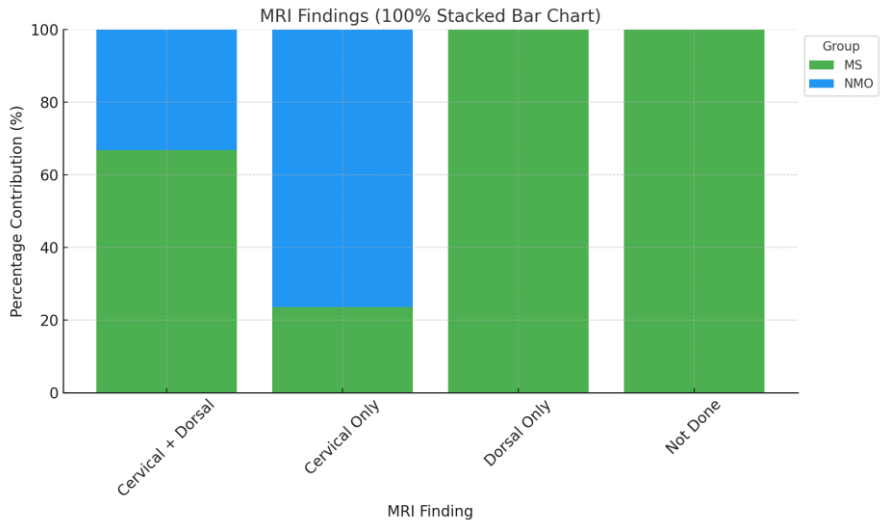

Figure 1: MRI Findings Comparison (Heat map)

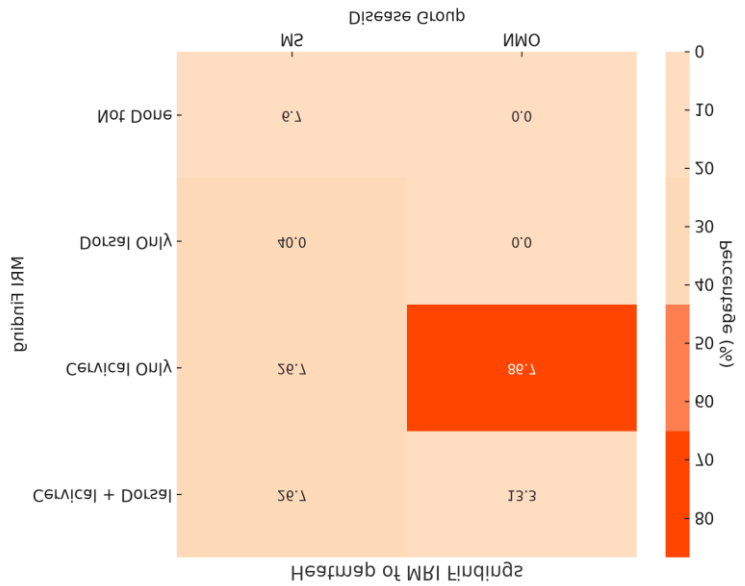

Figure 2: Detailed MRI Findings: MS vs. NMO

Cronbach's Alpha analysis

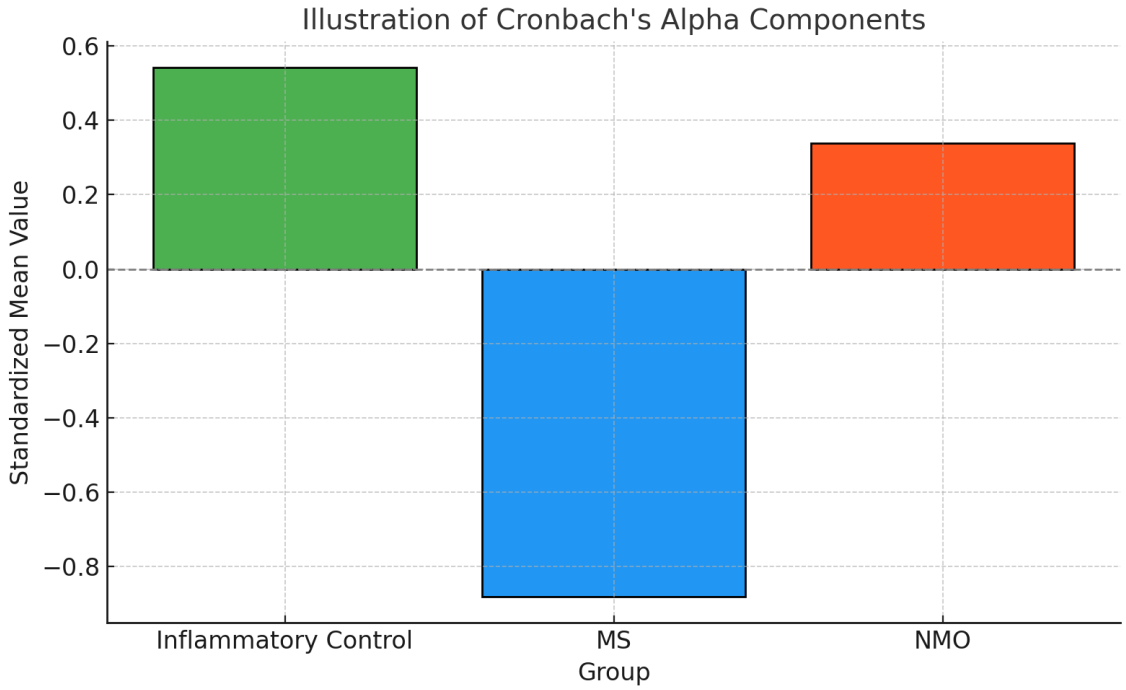

Figure 3: Standardized Mean Values Across Groups

Note: For the dataset on 'Disease Onset According to Age' across the groups: Inflammatory Control, MS, and NMO. The computed Cronbach's Alpha is 0.49, indicating moderate reliability. Below is the visual representation of the standardized mean values.

Discussion

Neuromyelitis optica spectrum disorder (NMOSD) typically manifests in the third or fourth decade of life and exhibits a strong female predominance. Our study aligns with global findings, as 90% of NMOSD cases occurred in females, and the mean age was comparable across all groups, corroborating the results of Lopez et al. (2021).

The average age of onset was 32.9 years for NMOSD, 32.67 years for inflammatory disorders, and slightly lower at 30.13 years for multiple sclerosis (MS). These figures are in agreement with the observations of Prasad et al. (2023) and Salman et al. (2022), who report early adulthood as the typical period for disease emergence across these conditions.

Interestingly, over half (53.3%) of our NMOSD patients had a monophasic disease course, contrasting with the commonly relapsing nature of NMOSD described in the literature. Akman-Demir et al. (2011) suggest that such discrepancies may result from methodological differences, including shorter follow-up durations or the inclusion of newly diagnosed cases. Meanwhile, the relapsing-remitting pattern, which is the hallmark of MS according to Myhr et al. (2001), was observed in only 33.3% of MS cases in our cohort, possibly reflecting variability in disease progression or access to treatment.

Visual impairment was a universal feature among NMOSD patients, reaffirming the prominence of optic neuritis in this condition, as highlighted by Wingerchuk et al. (1999). Additionally, walking difficulties were prevalent, affecting 93.3% of NMOSD patients and 73.3% of those with MS, indicating significant motor involvement consistent with previous findings (Stansbury, 1949).

Magnetic resonance imaging (MRI) findings further emphasized diagnostic distinctions: longitudinally extensive transverse myelitis (LETM), particularly involving the cervical spine, was present in 86.7% of NMOSD cases. In contrast, 40% of MS patients exhibited dorsal spinal cord lesions, supporting established imaging differences between the two conditions (Lennon et al., 2004; O'Riordan et al., 1996).

Cerebrospinal fluid (CSF) analysis revealed elevated protein levels without oligoclonal bands in more than half of NMOSD patients. In contrast, oligoclonal bands were detected in 96% of MS cases—a classic finding consistent with MS pathology (Bergamaschi et al., 2004).

The presence of AQP4-IgG antibodies in 86.7% of NMOSD patients in our study aligns with international prevalence rates and underscores the test's diagnostic specificity. The absence of these antibodies in most MS patients reinforces its utility in distinguishing NMOSD (Cabrera-Gómez et al., 2009; Pittock et al., 2008). Interestingly, only 13.3% of NMOSD patients tested positive for antinuclear antibodies (ANA), a lower rate than reported in other regions, which may suggest population-based immunogenetic variability (Nakashima et al., 2006).

Regarding treatment, 70% of NMOSD patients received a combination of steroids and immunosuppressants, possibly contributing to the monophasic course observed. Notably, plasma exchange therapy—known for its efficacy in acute attacks—was not used in any case, highlighting a treatment gap (Shimizu et al., 2007). In the MS group, 40% were managed with steroids alone despite their limited long-term efficacy in MS (National Collaborating Centre for Chronic Conditions, 2004).

Conclusion

This study highlights the diagnostic value of AQP4-IgG testing in distinguishing NMOSD from MS, especially in resource-limited settings such as Sudan. However, the limited availability of diagnostic tools and the absence of standardized national guidelines likely contribute to underdiagnosis and delayed treatment. There is a pressing need to enhance clinical awareness and establish clear diagnostic protocols to ensure timely and accurate identification of NMOSD.

Despite offering important insights, this study is limited by a small sample size and the lack of long-term follow-up data. Future research should focus on larger patient cohorts and investigate the clinical and therapeutic implications of AQP4-IgG positivity within the Sudanese population.

Reference :

1. Ahmed, A., Ali, B., Hassan, C., Mohamed, D., & Osman, E. (n.d.). Anti-aquaporin 4 IgG (NMO–IgG) antibodies among Sudanese patients with Neuromyelitis Optica and multiple sclerosis.
2. Chan, K. (2017). Immunopathological Effects of Aquaporin-4 Ig G in Neuromyelitis Optica Spectrum Disorders. *MOJ Immunology, 5*(5). MedCrave Group. https://doi.org/10.15406/moji.2017.05.00168
3. Cortese, R., Prados, F., Tur, C., Bianchi, A., Brownlee, W., Angelis, F. D., Paz, I. D. L., Grussu, F., Haider, L., Jacob, A., Kanber, B., Magnollay, L., Nicholas, R., Trip, A., Yiannakas, M., Toosy, A., Hacohen, Y., Barkhof, F., & Ciccarelli, O. (2022). Differentiating Multiple Sclerosis From AQP4-Neuromyelitis Optica Spectrum Disorder and MOG-Antibody Disease With Imaging. *Neurology, 100*(3). Lippincott Williams & Wilkins. https://doi.org/10.1212/wnl.0000000000201465
4. Jarius, S., Aboul-Enein, F., Waters, P., Kuenz, B., Hauser, A. E., Berger, T., Lang, W., Reindl, M., Vincent, A., & Kristoferitsch, W. (2008). Antibody to aquaporin-4 in the long-term course of neuromyelitis optica. *Brain, 131*(11), 3072. Oxford University Press. https://doi.org/10.1093/brain/awn240
5. Kim, H. J., Paul, F., Lana–Peixoto, M. A., Tenembaum, S., Asgari, N., Palace, J., Klawiter, E. C., Sato, D. K., Sèze, de, Wuerfel, J., Banwell, B., Villoslada, P., Saiz, A., Fujihara, K., Kim, S., Paul, F., Wuerfel, J., Cabre, P., Marignier, R., … O'Connor, K. C. (2015). MRI characteristics of neuromyelitis optica spectrum disorder. *Neurology, 84*(11), 1165. Lippincott Williams & Wilkins. https://doi.org/10.1212/wnl.0000000000001367
6. Akman-Demir, G., et al. (2011). Prognostic implications of aquaporin-4 antibody status in neuromyelitis optica patients. *Journal of Neurology, 258*(3), 464-470.
7. Bergamaschi, R., et al. (2004). Oligoclonal bands in Devic's neuromyelitis optica and multiple sclerosis. *Multiple Sclerosis, 10*(1), 2-4.
8. Cabrera-Gómez, J.A., et al. (2009). Neuromyelitis optica-positive antibodies confer a worse course in relapsing neuromyelitis optica. *Multiple Sclerosis, 15*(7), 828-833.
9. Lennon, V.A., et al. (2004). A serum autoantibody marker of neuromyelitis optica: distinction from multiple sclerosis. *The Lancet, 364*(9451), 2106-2112.
10. Lopez, J.A., et al. (2021). Pathogenesis of autoimmune demyelination: from multiple sclerosis to neuromyelitis optica spectrum disorders and myelin oligodendrocyte glycoprotein antibody-associated disease. *Clinical & Translational Immunology, 10*(7), e1316.
11. Myhr, K.M., et al. (2001). Disability and prognosis in multiple sclerosis. *Multiple Sclerosis, 7*(1), 59-65.
12. Nakashima, I., et al. (2006). Clinical and MRI features of Japanese patients with multiple sclerosis positive for NMO-IgG. *Journal of Neurology, Neurosurgery & Psychiatry, 77*(9), 1073-1075.
13. National Collaborating Centre for Chronic Conditions. (2004). *Multiple Sclerosis: National Clinical Guideline for Diagnosis and Management in Primary and Secondary Care.* London: Royal College of Physicians.

14. O'Riordan, J., et al. (1996). Clinical, CSF, and MRI findings in Devic's neuromyelitis optica. *Journal of Neurology, Neurosurgery & Psychiatry, 60*(4), 382–387.
15. Pittock, S.J., et al. (2008). Neuromyelitis optica and non-organ-specific autoimmunity. *Archives of Neurology, 65*(1), 78–83.
16. Prasad, C.B., et al. (2023). Overlap syndrome of anti-aquaporin four positive neuromyelitis optica spectrum disorder and primary Sjögren's syndrome. *Rheumatology International,* 1-9.
17. Salman, M.M., et al. (2022). Emerging roles for dynamic aquaporin-4 subcellular relocalization in CNS water homeostasis. Oxford University Press.
18. Shimizu, J., et al. (2007). Low-dose corticosteroids reduce relapses in neuromyelitis optica. *Multiple Sclerosis, 13*(8), 968-974.
19. Stansbury, F.C. (1949). Neuromyelitis optica; a review of literature. *Archives of Ophthalmology, 42*(3), 292–305.
20. Wingerchuk, D.M., et al. (1999). The clinical course of neuromyelitis optica (Devic's syndrome). *Neurology, 53*(5), 1107-1114.
21. Chan, K.H. (2017). Pathophysiology of Neuromyelitis Optica Spectrum Disorders: Beyond Aquaporin-4-IgG. *Frontiers in Neurology, 8,* 1–10.
22. Cortese, R. et al. (2022). Diagnostic and Treatment Advances in Neuromyelitis Optica Spectrum Disorders. *Nature Reviews Neurology, 18,* 543–558.
23. Jarius, S., & Wildemann, B. (2010). The history of neuromyelitis optica: Part 1 – Concepts of NMO before the Discovery of AQP4-IgG. *Journal of Neuroinflammation, 7,* 28.
24. Kim, S.H., et al. (2020). Mechanisms and Treatment of Neuromyelitis Optica Spectrum Disorder. *Journal of Clinical Neurology, 16*(1), 1–10.
25. Lennon, V.A., et al. (2004). A Serum Autoantibody Marker of Neuromyelitis Optica: Distinction from Multiple Sclerosis. *The Lancet, 364*(9451), 2106–2112.
26. Lennon, V.A., et al. (2005). IgG Marker of Neuromyelitis Optica: Specificity for AQP4 and Implications for Pathogenesis. *The Lancet Neurology, 4*(5), 371–377.
27. Marignier, R., et al. (2021). AQP4-IgG in Neuromyelitis Optica Spectrum Disorders: Clinical Correlations. *Neurology Neuroimmunology & Neuroinflammation, 8*(6), e1024.
28. Papadopoulos, M.C., & Verkman, A.S. (2013). Aquaporin-4 and Neuromyelitis Optica. *The Lancet Neurology, 12*(6), 535–544.
29. Pittock, S.J., et al. (2019). Eculizumab in AQP4-IgG–Positive Neuromyelitis Optica Spectrum Disorder. *New England Journal of Medicine, 381*(7), 614–625.
30. Saiz, A., et al. (2018). AQP4 Antibodies in Neuromyelitis Optica: Diagnostic and Prognostic Implications. *Neurology, 91*(13), 582–590.
31. Verkman, A.S., et al. (2014). Role of Aquaporins in Neuromyelitis Optica Spectrum Disorders. *Nature Reviews Neurology, 10,* 493–505.
32. Wingerchuk, D.M., et al. (2007). Neuromyelitis Optica: Clinical Features, Immunopathogenesis, and Treatment. *The Lancet Neurology, 6*(9), 805–815.
33. Wingerchuk, D.M., et al. (2015). Revised Diagnostic Criteria for Neuromyelitis Optica Spectrum Disorders. *Neurology, 85*(2), 177–189.
34. Yamamura, T., et al. (2019). Efficacy and Safety of Satralizumab in Neuromyelitis Optica Spectrum Disorder: Results from Two Phase 3 Trials. *The Lancet Neurology, 18*(12), 1089–1099.

Author Contributions

- *Marwa Alamin* (1): Led the study design, data collection, and primary manuscript drafting.
- *Malaz Alamin* (2): Contributed to data interpretation, literature review, and critical revision of the manuscript.
- *Anas Ahmed* (3): Performed statistical analysis and supported methodology development.
- *Sharif Ahmed* (4): Assisted in clinical evaluation of cases and contributed to the discussion section.
- *Rayan Morad* (5): Participated in data acquisition and ensured quality control throughout the study.
- *Wafaa Ali* (6): Reviewed all drafts for intellectual content and provided final approval for publication.

All authors read and approved the final manuscript.