ENCEPHALITIS CONFERENCE

2024

Poster Booklet

2ND – 3RD DECEMBER
ROYAL COLLEGE OF PHYSICIANS, LONDON AND VIRTUALLY

KEYNOTE SPEAKERS:

- **Professor Michael Wilson**
  UCSF Weill Institute for Neurosciences, USA
  "Metagenomics, Encephalitis and Global Health"

- **Dr Marianna Spatola**, University of Barcelona and La Caixa Research Institute, Spain
  "Functional Effects of Antibodies in Infectious and Autoimmune Encephalitis"

GUEST SPEAKERS:

- **Dr Ava Easton**, Encephalitis International & University of Liverpool, UK
  "Brains on Fire: Patient Outcomes and Quality of Life Following Encephalitis"

- **Dr Bhagteshwar Singh**, University of Liverpool & Royal Liverpool University Hospital, UK
  "Improving Diagnosis and Management of People with Brain Infections in Brazil, India and Malawi"

Encephalitis International
The brain inflammation non-profit

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Dr Afa Ibrahim

Nottingham University Hospital, UK

Comparison of Outcomes in Seropositive Autoimmune Encephalitis Presenting with Cognitive Dysfunction at Disease Onset: A Retrospective Audit at a Tertiary Neuroscience Centre

Dr Afa Ibrahim, from the Maldives, is in her final year of internal medicine training at Nottingham University Hospital Trust, with a dedicated ambition to specialise in neurology. A graduate of the prestigious All India Institute of Medical Sciences (AIIMS), India, she brings a robust academic foundation to her clinical and research pursuits. Dr Ibrahim's interests focus on neuroinflammatory and neuromuscular disorders.

Nottingham University Hospital NHS Trust

Encephalitis International The brain inflammation non-profit

Comparison of Outcomes in Seropositive Autoimmune Encephalitis Presenting with Cognitive Dysfunction at Disease Onset

A Retrospective Audit at a Tertiary Neuroscience Centre

Afa Ibrahim, Beili Shao, Crystal Teoh, Hina Khan, Akram Hosseini, Bruno Gran, Radu Tanasescu

1 INTRODUCTION

Cognitive dysfunction is a common and debilitating hallmark of seropositive autoimmune encephalitis (SPAIE), manifesting at any stage in the disease course and significantly contributing to morbidity and mortality. Literature on the impact of cognitive dysfunction during the early phase of the disease is limited, prompting us to conduct this study to better understand this aspect of the disease.

2 METHODS

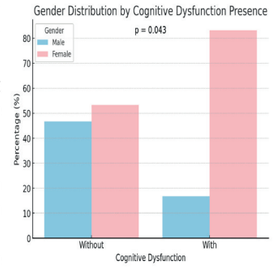
A retrospective audit(22-578C NUH) of 41 patients diagnosed with SPAIE from June 1995 to July 2023 identified 26 patients (63.41%) with cognitive dysfunction at onset and 15 patients (36.59%) without. Data were collected from electronic medical records on demographics, clinical presentation, investigations, treatment modalities, and outcomes, and were compared using univariate analysis. Functional outcomes were assessed with the modified Rankin Scale (mRS) at onset, 6-months, 12-months, and 1-year, and clinical outcomes were measured using Clinical Assessment of Severity in Encephalitis (CASE) scores at onset and last follow-up.

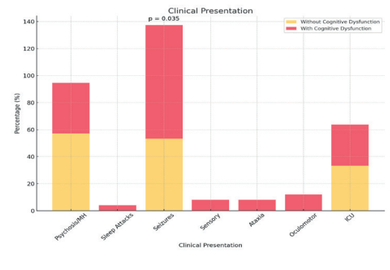
3 RESULTS

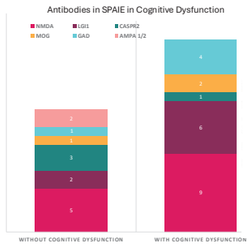
Demographics: Mean age was similar in both groups, with a predominance of females, especially in the cognitive dysfunction group.
Presenting Symptoms: Common: Seizures and psychosis, along with cognitive dysfunction. Seizure prevalence: Higher in the cognitive dysfunction group (80.77% vs 53.33%).

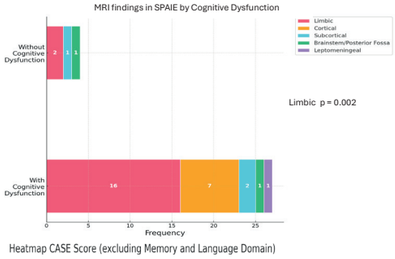
Antibodies: NMDAr antibodies were the most common in both groups, with similar rates. LGI-1 antibodies were more frequent in the cognitive dysfunction group (23.08% vs 13.33%).
MRI Findings: Limbic involvement was more prominent in the cognitive dysfunction group.

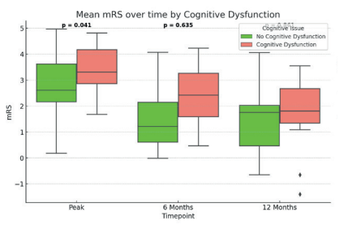
Treatment: Similar use of antiseizure medications and treatment approaches in both groups.
Outcome: Patients with cognitive dysfunction had a higher rate of poor functional status at onset (mRS 3-6: 85% vs 53%, p=0.029).
Worse scores in other CASE domains (excluding memory/language) for the cognitive dysfunction group.
Both groups saw favourable mRS (0-2) and excellent CASE scores (0-4) over time. The cognitive dysfunction group continued to have poorer outcomes overall. The most significant mRS improvement occurred within the first 6 months in both groups.

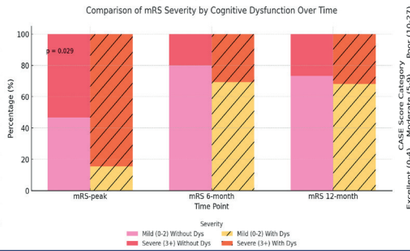


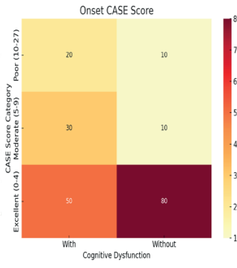


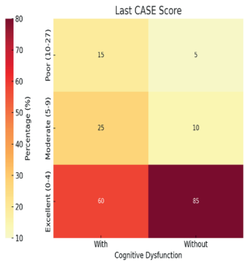












4 CONCLUSION

Patients with **SPAIE who present with cognitive dysfunction at onset** exhibit a distinct clinical phenotype, characterized by

- **Higher frequency of seizures**
- **Increased LGI-1 antibody positivity**
- **Greater limbic involvement on MRI**
- **Worse functional and clinical outcomes**

  mRS and CASE scores at disease onset and

  Substantial morbidity despite improvement with treatment.

- Cognitive dysfunction is typically reported in 80-90% of autoimmune encephalitis; however, our audit identified it in only 65%. This discrepancy may be explained by potential underdiagnosis in less severely ill patients.
- Within the limitations of our study, cognitive dysfunction emerges as an adverse prognostic indicator.
- Further research utilizing more comprehensive memory assessment tools is needed to better understand the patterns of cognitive dysfunction and guide therapeutic strategies.

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Steroid-Responsive Hashimoto's Encephalopathy in a 98 Year Old: A Case Report and Literature Review

Dr Alfeo Julius Sy is an adult neurology resident from Manila, Philippines, currently in his final year of training. Coming from a resource-limited country, Dr Sy is a highly motivated individual with a deep-seated curiosity to learn from global experts and integrate diverse perspectives into his practice. As a novice researcher, he is committed to advancing his skills, contributing to innovative research, and expanding his knowledge base. His enthusiasm for neurology is driven by a passion for continuous learning and a commitment to patient-centred care.

Steroid-responsive Hashimoto's encephalopathy in a 98 year old: a case report and systematic review

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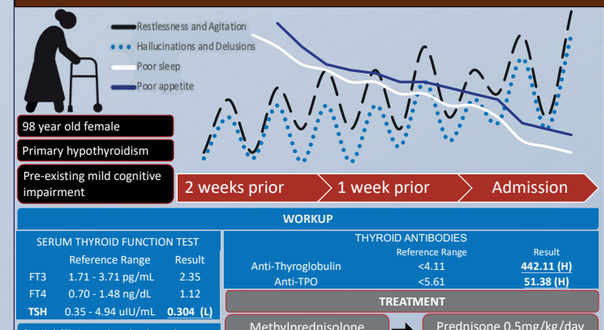
INTRODUCTION

- Hashimoto's encephalopathy (HE) is a rare autoimmune disorder under the umbrella category of steroid-responsive encephalopathies[1], and can present with a variety of neurologic symptoms from focal deficits to global confusion.
- In cognitively intact individuals, late-life neuropsychiatric symptoms may be seen as preclinical and prodromal symptoms of dementia and its subtypes[2].
- A high index of suspicion is required to diagnose HE due to a lack of well-defined criteria, often leading to a diagnosis of exclusion. Currently, the most recognized diagnostic criteria for HE require the presence of (a) encephalopathy with hallucinations, seizures, stroke-like episodes, or myoclonus, (b) elevated serum thyroid antibodies, (c) subclinical or mild overt thyroid disease, (d) normal or nonspecifically abnormal cranial imaging, and the absence of antineuronal antibodies in the serum or CSF, and (f) reasonable exclusion of an alternative diagnosis[3].
- Nonetheless, it is an important diagnosis to consider as early initiation of treatment results in good outcomes and must be considered in cases of "investigation negative encephalopathies" [4].
- We present a case of a 98 year old female with pre-existing mild cognitive impairment presenting with psychotic symptoms and elevated serum thyroid antibodies successfully treated with a reduced dose of steroids; and review the clinical characteristics, diagnostic challenges, and management outcomes of new onset HE in this unique and vulnerable population.

CASE PRESENTATION



METHODS

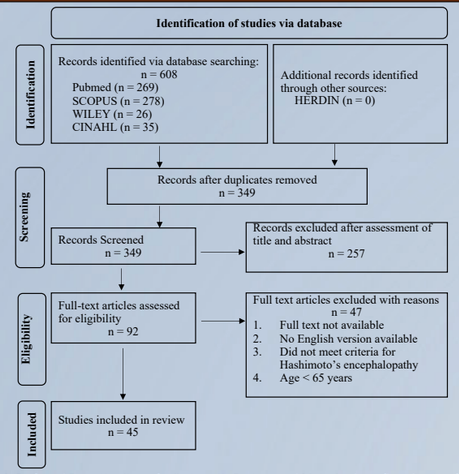


Figure 1. Preferred Reporting Items for Systematic Reviews and Meta-Analyses flow diagram for this review.

RESULTS

Diagnostic tests	Findings (n = 52)
Serum thyroid function test	
Within normal limits	25 (48.0%)
Elevated TSH, no data on FT3 & FT4	16 (30.7%)
Elevated TSH, low FT3, normal FT4	2 (03.8%)
Low TSH, normal FT3 & FT4	2 (03.8%)
Low TSH, no data on FT3, Low FT4	1 (01.9%)
Low TSH, elevated FT3 & FT4	1 (01.9%)
No data presented	5 (09.6%)
Serum thyroid antibodies	
Elevated Tg-Ab, elevated anti-TPO	41 (78.8%)
Elevated Tg-Ab, normal anti-TPO	5 (09.6%)
Normal Tg-Ab, elevated anti-TPO	6 (11.5%)
Cerebrospinal fluid analysis	
Elevated protein	25 (48.0%)
Unremarkable	17 (32.6%)
No data presented	10 (19.2%)
Electroencephalogram	
Generalized or focal slowing	32 (61.5%)
Focal spikes and discharges	9 (17.3%)
Within normal limits	6 (11.5%)
No data presented	5 (09.6%)
Initial steroid dose	**Findings and outcome (n = 52)**
Methylprednisolone (IV)	
1 gm/day	19 (36.5%) Good outcome (9/19) Partial Response (5/19) No response (2/19) Expired (2/19)
500 mg/day	6 (11.5%) Good outcome (5/6) Partial Response (1/6)
1 mg/kg/day	5 (09.6%) Good outcome (4/5) No response (1/5)
Dexamethasone (IV)	
24 mg/day 16 mg/day	1 (01.9%): Good outcome (1/1) 1 (01.9%): Good outcome (1/1)
Oral Prednisone varying doses, duration, and tapering	9 (17.3%) Good outcome (4/9) Partial Response (4/9) No response (1/9)
Unspecified steroid formulation and dose	5 (09.6%)

Table 1. Pertinent diagnostic tests, treatment regimen, and outcomes in older patients with new onset HE. TSH, thyroid stimulating hormone; FT3, free triiodothyronine; FT4, free thyroxine (FT4); Tg-Ab, antithyroglobulin antibodies; anti-TPO, thyroid peroxidase antibodies

DISCUSSION

- The clinical presentation of HE in older persons can be heterogeneous. It often mimics other neurological or psychiatric conditions including neurodegenerative diseases, leading to diagnostic challenges and delays in management. Moreover, older individuals often have comorbidities and are on multiple medications, which can complicate the clinical picture[4].
- In a setting of rapidly progressing dementia, normal brain imaging, and normal routine diagnostics, HE should be suspected[5].
- Treatment of HE is based on high-dose steroids with subsequent tapering, but there are currently no specific guidelines and treatment protocols. Intravenous methylprednisolone is used in most cases, while the dose and length of administration remains to be based on the physician's clinical reasoning and patient's response to therapy.
- Most patients respond to treatment after a short course, and some can take up to the first 4 to 6 weeks for clinical improvement to be noticeable[6].
- A lower dose of steroids (Methylprednisolone 500 mg/day intravenously) may be a reasonable choice for patients with weak immune systems and a higher risk of infections.

CONCLUSION

- HE is a rare but potentially reversible cause of encephalopathy in the elderly.
- In an elderly patient with preexisting mild cognitive impairment and rapidly progressing neuropsychiatric symptoms, HE should be suspected.
- Steroids remain to be the mainstay of treatment, but a nuanced approach is required in the older population, balancing the benefits of immunomodulatory therapies with potential risks, and ensuring optimal thyroid hormone replacement therapy.
- Our patient to our knowledge is the oldest diagnosed case of new onset HE treated successfully with steroids at a reduced dose.
- Further research is needed to better understand the prevalence, diagnosis, and management of HE in the older population, which can help improve patient outcomes in this vulnerable population.

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Dr Anlys Olivera

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Healthy skepticism: trial of immunosuppression in treatment resistant psychosis, a case report

Dr Anlys Olivera is a resident in the Neurology and Psychiatry Residency Program at New York University Langone Medical Center in New York City. Most of their research work has focused on the neuropsychiatric symptoms of neuroimmune processes specially in brain injury.

Healthy skepticism: trial of immunosuppression in treatment resistant psychosis, a case report

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ABSTRACT

Up to 30% of individuals with a psychotic disorder such as schizophrenia meet criteria for treatment resistance. When clozapine or ECT fails to show a benefit, all hope may seem lost, and these patients are often institutionalized,. Sometimes the clinical suspicion for a non-psychiatric etiology is high enough that a basic workup is completed, but if there are no clear abnormal findings suggestive of an autoimmune/paraneoplastic process, then the likelihood of being trialed on immune based therapy is nonexistent. Here we present a case of a women who developed acute onset psychosis and mania with significant cognitive deficits. These symptoms were refractory to commonly used treatment modalities in psychiatry for over a year. Significant improvements in symptom burden and return to functional status was achieved only after starting high dose steroids and IVIG treatment. This case highlights some of the challenges of diagnostic uncertainty and pursuing alternative therapeutic interventions in treatment resistant psychosis.

INTRODUCTION

- Criteria for treatment resistant psychosis: failure to respond to two antipsychotic trials with a therapeutic dose and adequate duration[1]
- Evidence for immune dysregulation in schizophrenia and support for an inflammatory component to treatment resistant psychosis[2,3]
- Psychosis as the most common manifestation of AE and “Red Flags” in the diagnosis of a primary psychotic disorder[4]
- Antibody-negative AE and decision to treat[5,6]
- Differential diagnosis of AE with psychiatric features

CASE

- 37yo woman, from Central America, married, domiciled with husband and two daughters (3yo, 16yo), employed, no significant past medical history, no prior psychiatric history. No known family history of autoimmune/inflammatory conditions or malignancy. No toxic habits.
- The day prior to symptom onset patient reported generalized malaise, then family describes acute onset psychosis, followed by manic symptoms, cognitive deficits, and disorientation that progress without remission until initial presentation to the hospital in mid November 2022. Vitals notable for persistent tachycardia to low 130s. Basic labs notable for COVID+, thrombocytosis, and an anion gap, with a head CT that was unrevealing.

CASE CONTINUED

- Patient admitted to psychiatry, and over the next 9 months she is repeatedly cycling between symptoms of psychosis (ex. auditory hallucinations, paranoid ideations, delusions), mania (ex. insomnia, poor attention, impulsive, grandiose, sexually preoccupied), severe depression (ex. withdrawn, dysphoric, psychomotor slowing), and catatonia (including periods of excited/agitated catatonia and stuporous catatonia), with cognitive impairments (ex. short term memory deficits, executive dysfunction, attention deficits, disorientation, inability to recognize family). During this period patient was trialed on multiple antipsychotics, mood stabilizers, antidepressants, anxiolytics, sleep aids, amantadine and memantine, with little to no benefit. She started ECT in May 2023, and in late August she is discharged home on Quetiapine, Lorazepam, Lamotrigine, Escitalopram, Synthroid, and outpatient ECT.
- Patient returns to emergency department in mid November 2023, with acute on chronic decompensation. She is admitted to psychiatry, continued on ECT and started on clozapine. No sustained improvements reported by late January 2024, with application initiated requesting a transfer to state hospitals.
- Work up: during initial hospitalization she is diagnosed with papillary thyroid carcinoma. TSH 5.39, FT4 0.7, thyroglobulin 104, anti-TPO/thyroglobulin Ab negative. Underwent partial lobectomy in August 2023. Her labs show positive ANA (1:320) with negative ANA-ENA panel, C3/C4 within normal limits, CRP 122, ESR 83, SPEP w/ elevation in alpha 2 region. HIV/RPR/hepatitis labs were negative. Normal levels of vitamin B1/B6/B12. Copper/ceruloplasmin and heavy metal screen labs unremarkable. Paraneoplastic serum panel was positive for VGCC P/Q. ENS2 panel negative, NMDA R Ab in serum negative. CSF studies from March 2023 and January 2024, with normal protein, cell count, and glucose, a negative ME panel, negative CSF cultures. 14-3-3 and T-tau within normal limits, RT-QuIC negative. ENC2 panel negative. MRI brain with contrast in December 2022, and a repeat in June 2023, and March 2024, did not show any abnormalities. A routine EEG in January 2023 did not show focal abnormality/epileptiform discharges. No other e/o malignancy on CT chest/abdomen/pelvis.
- End of January 2024, patient starts high dose steroids, followed by a course of IVIG in early February 2024. Pt discharged to home in early March 2024, on Lorazepam taper, steroid taper, clozapine, and lamotrigine, with plan to taper ECT sessions outpatient (ended in October). She received 3 courses of monthly IVIG between July and September 2024. As of November 2024, patient continues to progress towards new baseline, fully functional at home, planning to return to work by end of year, with persistent personality changes.

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DISCUSSION

- The abrupt onset of psychotic/manic symptoms with significant cognitive deficits (with paroxysmal spells) in setting of COVID infection and new diagnosis of papillary thyroid cancer, and symptoms refractory to multiple treatment modalities in psychiatry including clozapine and ECT (with absence of risk factors for treatment resistant psychosis) does not represent a typical illness script for a primary psychotic/mood disorder
- No guidelines on what to do when labs/imaging does not support any one specific diagnosis. The significant improvements in mental status and functional status after steroids and IVIG that have been sustained despite tapering psychotropic meds, is suggestive of immune process contributing to symptoms. No guidelines for maintenance treatment
- Limitations of diagnosing psychotic disorders: spectrum of clinical syndromes without clearly defined pathophysiology/biomarkers
- Limitations of diagnosing AE/paraneoplastic process: incomplete CSF workup, limited neoplasm workup, limitations of imaging modalities, lack of neuropsychological testing
- Implications for anchoring biases
- Ethics of trialing immunosuppressive therapies in absence of supporting objective data
- Specific challenges with coordinating interdisciplinary care in this population

CONCLUSIONS

- What happens next?: no guidance on prognosis, how to monitor progress, or recommendations for maintenance therapies
- Importance of encouraging a healthy skepticism when evaluating patients in psychiatry and keeping a wide differential
- Need to clearly define “red flags” in this population that should trigger a workup for AE/paraneoplastic syndrome
- Developing guidelines for complete workup in this population
- Avoiding therapeutic inertia
- Developing guidelines to guide trial of immunosuppressive therapies in undifferentiated cases in psychiatry, and sequential escalation of immune therapies
- Ethical considerations in this population, and weighing the risk of doing nothing
- Neuropsychiatry as a liaison service

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Understanding the effects of anti-NMDA receptor autoantibodies on neurodevelopment using a human induced pluripotent stem cell-derived neurosphere assay.

Dr Babak Soleimani is a neurology registrar and Brain Entry Fellow based at the University of Oxford, working with the Oxford Neuroimmunology and Immunopsychiatry. Laboratory and Oxford Autoimmune Neurology Group. He is particularly interested in improving patient outcomes through understanding pathophysiological mechanisms of encephalitis and neuroimmune disease to help identify therapeutic targets.

Understanding the effects of anti-NMDA receptor antibodies on neurodevelopment using a human induced pluripotent stem cell-derived neurosphere assay.

Babak Soleimani[1,2], Jonathan Cleaver[2], Laurissa Havins[1,2], Gregory Holt[1,3], Annapoorna Kannan[1], Thomas Johnson[1], Sarosh Irani[2,4], Adam Handel[2] and Lahiru Handunnetthi[1,2,3].

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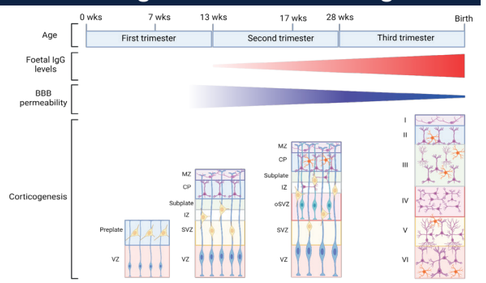
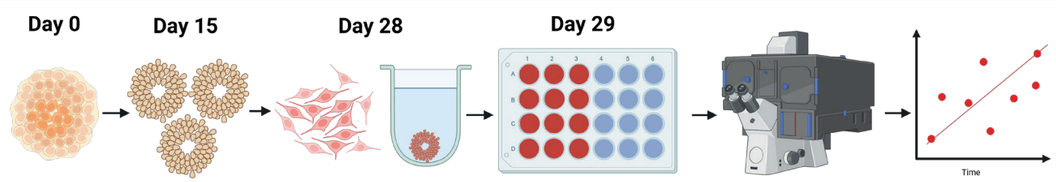
Background & Aims

- The transfer of antibodies from mother to her foetus is a vital protective immune mechanism. This maternal-to-foetal transfer begins around 13 weeks of gestation and IgG1 subclass is preferentially transferred across the placenta.
- N-methyl-D-aspartate receptor (NMDAR) encephalitis (NMDARE) predominantly affects young women of childbearing age.
- Anti-NMDAR antibodies are pathogenic and are predominantly subclass IgG1. Maternal-to-foetal transmission of these antibodies has been previously demonstrated[1].
- Animal studies highlight that anti-NMDAR antibodies can influence corticogenesis in mice, but the results are inconsistent. Importantly, no studies have examined effects of these antibodies on human neurodevelopment.
- Aim: To investigate the effects of anti-NMDAR antibodies on human corticogenesis using a human induced pluripotent stem cell (iPSC) neurosphere assay[2].

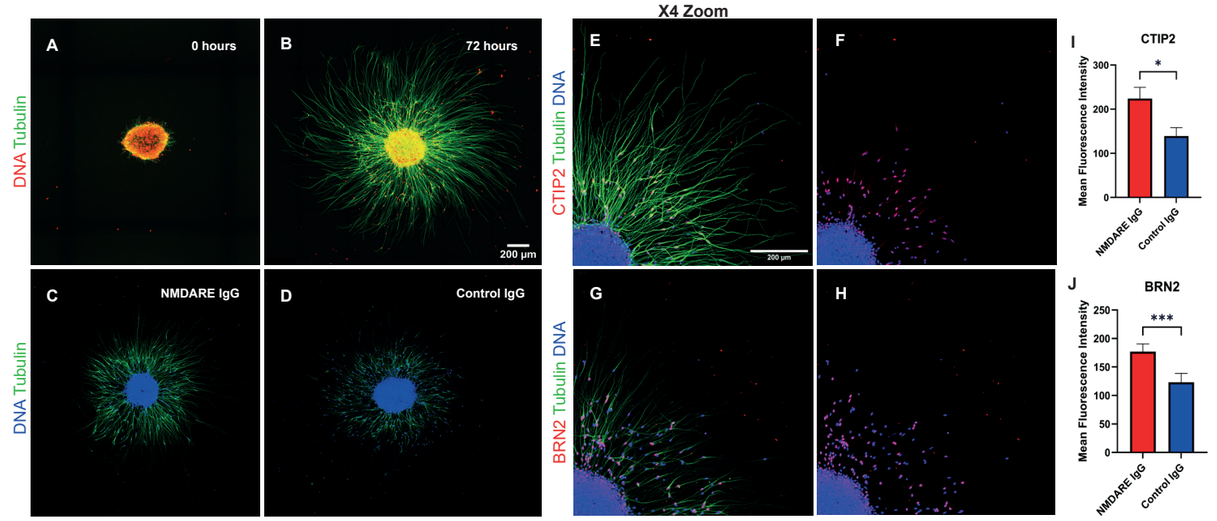
Maternal IgG transfer & Corticogenesis



Methods



Results



A and **B**: Live imaging of neurospheres at timepoints 0 and 72 hours. **C** and **D:** Fixed neurospheres at 72 hours following exposure to NMDARE and healthy control IgG respectively. **E - H**: CTIP2 (deep cortical layer) and BRN2 (superficial cortical layer) makers are expressed in neurospheres. **I** and **J**: Intensity analysis revealed significantly higher mean intensity of both CTIP2 (NMDARE IgG: 224.2±14.58, control IgG: 139.0±9.709, p=0.0101) and BRN2 (NMDARE IgG: 176.9±6.794, control IgG: 123.2±6.997, p=0.0009) markers in NMDARE IgG exposed neurospheres. Number of neurospheres assessed for CTIP2 marker: NMDARE IgG is n=3 and control IgG is n=4; and BRN2 marker: NMDARE IgG is n=4 and control IgG is n=5.

Discussion

- This study shows for the first time that anti-NMDAR antibodies can influence human cortical developmental processes.
- Further work is required to understand whether the observed anti-NMDAR antibody mediated changes to cortical development will lead to long-term consequences in the brain.
- This adds to the growing body of evidence that suggests maternal transfer of autoantibodies can alter neurodevelopmental processes. This carries implications for treatment strategies in this patient group during pregnancy.

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Dr Beili Shao

Neurology, Queen's Medical Centre, Nottingham University Hospitals NHS Trust, Nottingham, UK

Reappraisal of diagnosis in seronegative autoimmune encephalitis: 6-year experience of tertiary neuroscience centre

Dr Beili Shao is a specialist registrar and NIHR clinical lecturer in neurology with extensive academic and clinical training. She obtained her PhD in Clinical Neuroscience from the University of Nottingham in 2014, following her Masters in Neurology (2010) and Bachelors degree (2008) from Shanghai Jiaotong University. Dr Shao has professional experience spanning multiple institutions. Since 2019, she has been engaged in Neurology SpR training. Prior roles include core medical training (2016-2019) and a Junior Clinical Fellowship in Care of the Elderly at Royal Gwent Hospital (2015-2016). She holds MRCP SCE Neurology (2021) and MRCP PACES (2017) qualifications and has completed various advanced courses in multiple sclerosis, intensive care transfer, and Botox injections. Her research interests include the impact of hyperglycaemia on the blood-brain barrier, stroke, dementia, and multiple sclerosis. She has authored several peer-reviewed publications and is currently involved in clinical trials on myasthenia gravis and autoimmune encephalitis. Her work has been recognized with numerous awards, including poster prizes at various medical conferences. In addition to her clinical and research roles, Dr Shao is active in teaching and has supervised Masters students and delivered lectures to undergraduates and foundation doctors. She has also held several administrative roles, including Student Representative and Member of the Postgraduate Research Degrees Committee. Dr Shao's commitment to advancing medical science and patient care is evident in her ongoing research and clinical endeavours.

REAPPRAISAL OF DIAGNOSIS IN SERONEGATIVE AIE: 6-YEAR EXPERIENCE OF TERTIARY NEUROSCIENCE CENTRE

Beili Shao1,2, Crystal Teoh2, Afa Ibrahim2, Bruno Gran2, Akram A. Hosseini1,2, Radu Tanasescu2

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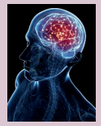
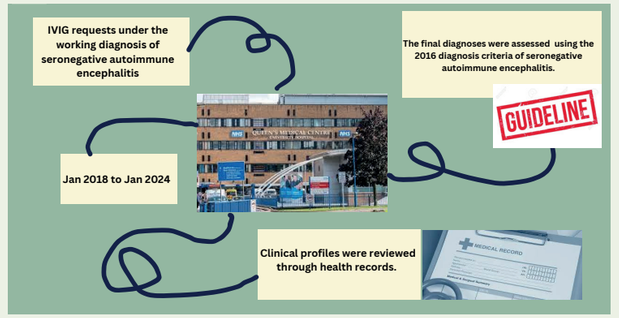
1 INTRODUCTION



Seronegative autoimmune encephalitis (SNAIE) is a spectrum of disorders often characterised by diagnosis challenges, delayed treatment and poor prognosis.

Intravenous immunoglobulin (IVIG), combined with intravenous methylprednisolone, is the first-line immunotherapy for severe autoimmune encephalitis. Accurate diagnosis is crucial to avoid unnecessary treatments and optimise patient outcomes.

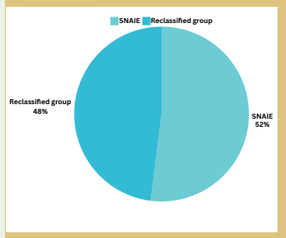
2 METHODS

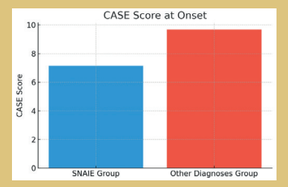


3 RESULTS

A. 48% of cases received IVIG had diagnosis reclassified.

25 cases of suspected SNAIE were identified, comprising 19 females and 6 males, with an average age of 36.36. 12 cases had diagnosis reclassified.





CASE Score was higher in reclassified group compared to the SNAIE group at onset.

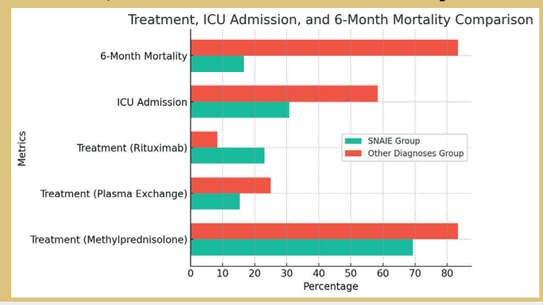
B. What are the alternate Diagnoses?

Alternate Diagnoses
Hashimoto encephalitis (1)
Mitochondrial disease (2)
Febrile infection-related epilepsy syndrome (2)
Post-Covid acute disseminated encephalomyelitis
Post-Covid leukoencephalopathy
Post-vaccine encephalomyelitis
Cerebral vasculitis
Lewy body dementia
Seronegative encephalitis (not meeting 2016 SNAIE criteria)
Status epilepticus (unknown cause)

C. Clinical presentation, MRI brain abnormalities and CSF findings

	SNAIE N=13	Reclassified group N=12
MRI Brain Abnormalities	61.54%	75%
Affected Area	Limbic	Cortical
OCB and cell count	1 OCB +	1 OCB + and 2 pleocytosis
Most common presentations	Cognitive issues, seizures, and psychosis	cognitive issues, seizures, and ataxia

D. Treatment, ICU admission and 6-m mortality



-IV MP was administered in a high proportion of cases in both groups
-Plasma exchange and rituximab were used in 15.38% and 23.08% of SNAIE cases, respectively, compared to 25% and 8.33% in the reclassified group.
-The six-month mortality rate was significantly higher in the other diagnoses group (83.33%) compared to the SNAIE group (16.67%) as well as ICU admission.

4 DISCUSSION

IVIG is a high cost treatment with associated risks, including thrombus and embolism. The high proportion of alternate diagnoses highlights the need for precise diagnostic protocols. The similarity in clinical presentations and investigation findings between SNAIE and other diagnoses underscores the complexity of SNAIE diagnosis. Enhanced diagnostic tools, such as advanced imaging techniques and novel biomarkers, may improve diagnostic accuracy.

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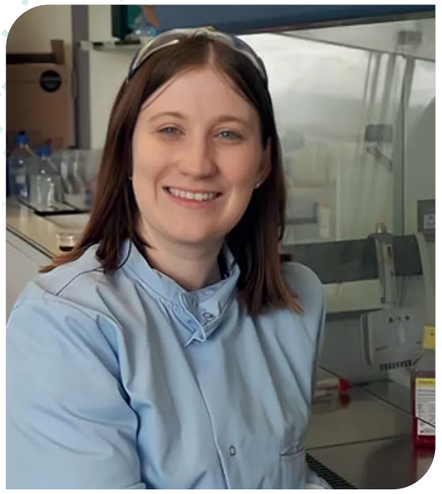

Dr Claire D. Hetherington

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In vitro modelling of seizure genesis during HSV-1 encephalitis

Dr Claire Hetherington is a postdoctoral research associate in Professor Benedict Michael's Infection Neuroscience lab. She began her scientific research career with a MBiolSci in Biochemistry & Molecular Cell Biology at the University of Sheffield, completing her masters research with Pfizer and Astrazeneca. She then undertook doctoral research in Translational Neuroscience at the University of Aberdeen, under the supervision of Dr Guy Bewick researching in vitro modelling of motor neurone disease using induced pluripotent stem cells (iPSCs). In 2023, she moved to the University of Liverpool to undertake postdoctoral research on in vitro modelling of CNS infections. Her research interests include in vitro models of neurological disease, stem cells, microfluidics and electrophysiology.

In vitro modelling of seizure genesis during HSV-1 encephalitis

Hetherington, C. D.[1], Nkongho F.[1], Dunai C.[1], Boardman S.A.[1],, Michael B.D.[1,2]

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[2]National Institute for Health Research Health Protection Research Unit in Emerging and Zoonotic Infections, Institute of Infection, Veterinary and Ecological Sciences, University of Liverpool, Liverpool, UK

The Infection Neuroscience Lab

UNIVERSITY OF LIVERPOOL

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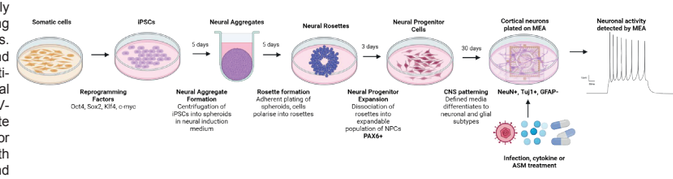
INTRODUCTION

The high rate of acute seizures and status epilepticus (SE) contributes to the severity and adverse clinical outcome of herpes simplex virus-1 encephalitis (HSE).[1] However, little is known about the specific mechanisms that influence the development of seizures and SE in patients.[1] Thus, there is a need for *in vitro* models using human cells to perform mechanistic studies and provide a translational platform for drug discovery. Two hallmarks of seizure generation are hyperexcitability of neurons and hypersynchrony of neural circuits.[2] Hyperexcitability is known to be induced by pro-inflammatory cytokines, of which astrocytes and microglia are a major source during infection.[3] We have developed an *in vitro* model of seizures provoked by HSV-1 infection using human iPSC-derived cortical neurons to identify mechanisms by which neuronal network hyperexcitability may be caused by HSV-1 infection; directly downstream of neurotropism or indirectly through local neuroimmune responses.

METHODOLOGY

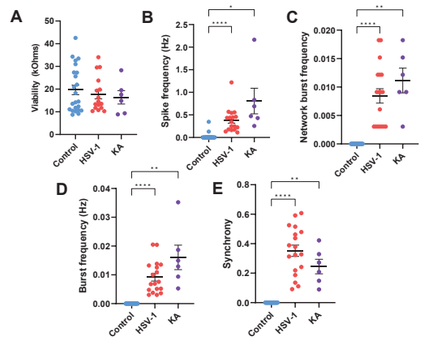
We have developed an in vitro model of seizures provoked by HSV-1 infection using human iPSC-derived cortical neurons (hiPSC-CNs) - differentiated using an established protocol (Karch et al, 2019, Figure 1) - to identify mechanisms by which neuronal network hyperexcitability may be provoked by HSV-1 infection through neurotropism or indirectly through pro-inflammatory cytokine secretion. This platform is also amenable to testing antiseizure medications (ASM) or immunomodulatory facots which may mitigate seizures. hiPSC-CNs were first validated using immunohistochemistry for neural progenitor and neuronal markers (data not shown). We then established functional hiPSC-CNs on a multi-electrode array which measures neuronal electrophysiological activity and viability in real time. hiPSC-derived cortical neurons and primary human astrocytes were infected with HSV-1 at varying MOIs, treated with previously infected (inactivated and filtered) astrocyte supernatant, treated with the seizuregenic compound kainic acid (KA) as a positive control or alternatively sham-infected with sterile Vero supernatant as a negative control. Both spontaneous and electrically evoked neuronal spike frequency, burst frequency and synchrony were non-invasively quantified at regular intervals before and after exposure using a Maestro Edge multi-electrode array (Axion Biosystems).



RESULTS

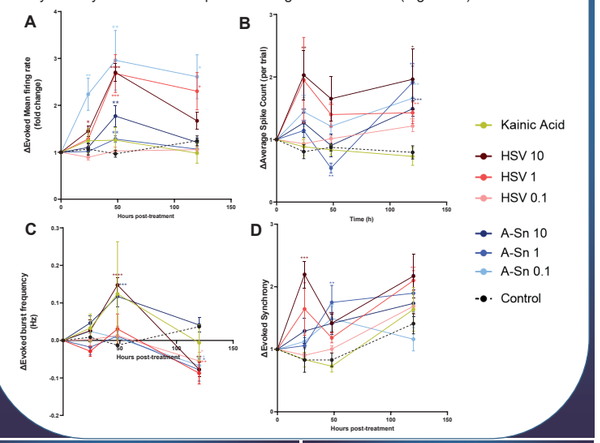
HSV-1 induces spontaneous hyperexcitability

hiPSC-CN cultures at 20 hours post-infection with HSV-1 KOS at an MOI of 0.1 showed no significant decrease in viability at this early timepoint compared to mock-infected (Fig 2a). Spontaneous spike frequency, burst frequency, network burst frequency, and synchrony in HSV-1 infected neurons were l significantly increased compared to mock-infected cells, with no significant difference between infected neurons and neurons treated with KA (Fig 2b-e).



HSV-1 induces evoked hyperexcitability

hiPSC-CN cultures were challenged with HSV-1 at MOIs of 10, 1 and 0.1 as well as exposure to inactivated, filtered supernatant from astrocytes infected for 24h with HSV-1 at MOIs of 10, 1 and 0.1. Increase or decrease from baseline in evoked (electrically stimulated) excitability parameters was calculated as a fold change from baseline for each well at regular time points up to 120h. Fold change in evoked mean firing rate was significantly increased compared to control at 24h, 48h, and 120h for HSV-1 infected and astrocyte supernatant (A-Sn) treated neurons (Fig 3a). Average spike count per stimulation trial was also increased in direct infection at 24-120h and after A-Sn stimulation at 24h and 120h (Fig 3b). Evoked burst frequency was also increased at 48h for high dose HSV and A-Sn conditions, comparable to KA, before reducing sharply by 120h (Figure 3c). Synchrony was also increased in high-dose infected (MOI 10) and A-Sn MOI 1 treatment at 24-120h with A-Sn showing a delayed onset of synchrony increase in comparison to high-dose infection (Figure 3d).



Conclusions

Our model assesses the electrophysiological activity of human neurons *in vitro*, producing seizure-like activity in response to infectious agents and drugs. Early-stage HSV-1 infection of hiPSC-CNs and exposure to supernatant from infected astrocytes causes the neuronal network to become hyperexcitable. Both HSV-1 infection and KA increase baseline spontaneous excitability. By contrast, HSV-1 infection increases spontaneous and evoked activity as does exposure to infected astrocyte supernatant, whereas KA does not, suggesting an alternative mechanism by which HSV-1 infection and local neuroimmune responses drive seizures.

FURTHER WORK

We are assessing the cytokine profile of infected astrocytes and iPSC-CNs and will identify potential targets to investigate whether they enhance or mitigate seizure activity *in vitro*. This is in tandem with a comparison of the cytokine profile in the CSF and serum of HSE patients who had seizures during their acute illness compared to those who did not. Finally, this powerful translational platform will be used to determine the ability of immunomodulatory therapies and anti-seizure medications to inhibit seizure-like activity *in vitro*.

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Dr Cordelia Dunai

CIMI, IVES, University of Liverpool, UK

Pulmonary SARS-CoV-2 infection leads to viral-independent para-infectious immune activation in the brain

Dr Cordelia Dunai is a postdoc research associate in Professor Benedict Michael's Infection Neuroscience lab. She is part of the Biomarkers and Immunology Working Group of the COVID-CNS study and the Liverpool Brain Infections Group. She earned her Ph.D. in immunology in 2021 in the lab of Professor William J. Murphy at UC Davis. Her interests include: immunology, virology, cancer biology, translational research, and scientific outreach..

Pulmonary SARS-CoV-2 infection leads to viral-independent para-infectious immune activation in the brain

Cordelia Dunai[1], Claire Hetherington[1], Sarah Boardman[1], Franklyn Egbe[1], Mark Ellul[1], Edward Needham[2], Jordan Clark[1], Parul Sharma[1], Kukatharmini Tharmaratnam[1], Yun Huang[1], Krishanthi Subramaniam[1], Robyn Williams[3], Ceryce Collie[1], Sophie Pendered[1], Greta Wood[1], Brendan Sargent[1], Geraint Sunderland[1], Arina Tamborska[1], Alexander Grundmann[4], Ian Galea[4], Alasdair Coles[2], Michael Griffiths[1], Tom Solomon[1], Gerome Breen[5], Sarosh Irani[3], Jonathan Cavanagh[6], Angela Vincent[3], Anja Kipar[7], James Stewart[1], Leonie Taams[2*], David Menon[2*], Benedict Michael[1*],

1. University of Liverpool; 2. University of Cambridge; 3. University of Oxford; 4. University of Southampton 5. King's College London; 6. University of Glasgow; 7. University of Zurich *joint senior authors

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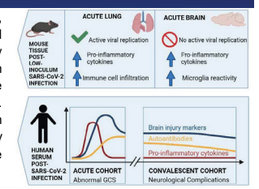
ABSTRACT

Although SARS-CoV-2 causes a respiratory viral infection, there is a significant incidence of neurological complications, including headache, seizure, encephalitis, and stroke, occurring in COVID-19 patients. Little is known about the likely diverse mechanisms causing these pathologies and there is a dire need to understand them and learn how to prevent and treat them. We measured brain injury markers and cytokines in the serum of study participants who had COVID-19. Brain injury markers (Nfl-L, UCH-L1, Tau, and GFAP) were measured by Quanterix Simoa and 48 cytokines were measured by Bio-rad Bioplex-200. We found that two brain injury markers and several cytokines/receptors were significantly associated with an abnormal Glasgow Coma Score in acute COVID-19 patients. For the mouse model of parainfectious effects on brain, 2-4 mo heterozygous K18-hACE2 transgenic C57BL/6 male and female mice were infected with $1x10^3$ PFU SARS-CoV-2. On day 5 post-infection, mice were euthanized and serum, brain, and lungs collected for RNA and protein analysis. We observed microglia reactivation in the brain of low dose-infected mice —interestingly, this was in the absence of active viral replication. Overall, we highlight two findings: (1) SARS-CoV-2 does not directly invade the brain in the majority of cases and so the associated neurological complications might arise from indirect effects, such as immune activation, which we can model in mice (2) although the immune system plays a critical role in controlling the virus, its dysregulation can cause pathology.



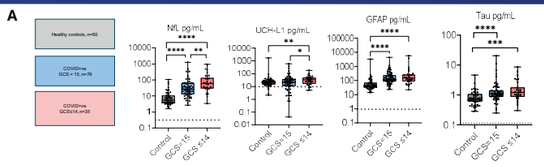
METHODS

We measured brain injury markers and cytokines in the serum of participants who had COVID-19 from the pandemic response study: ISARIC. We examined those participants with a normal vs. abnormal Glasgow Coma Score. In the COVID-CNS study, we could stratify by participants with neurological complications and specific pathologies. Brain injury markers (Nfl-L, UCH-L1, Tau, and GFAP) were measured by Quanterix Simoa and 48 cytokines were measured by Bio-rad Bioplex-200.

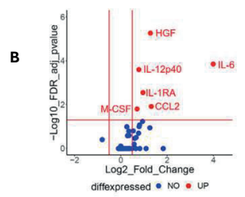
For the mouse model of parainfectious effects on brain, 2-4 mo heterozygous K18-hACE2 transgenic C57BL/6 male and female mice were infected intranasally with $1x10^3$ PFU or $1x10^4$ PFU SARS-CoV-2. On day 5 post-infection, mice were euthanized and serum, brain, and lungs collected for RNA and protein analysis.

RESULTS

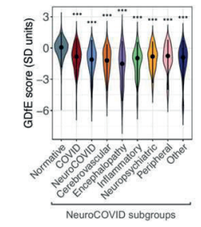
Biomarkers and long-lasting effects of COVID-19 on cognition in humans



Brain injury markers are highest in the ISARIC COVID group that had Glasgow Coma Scale (GCS) less than 15

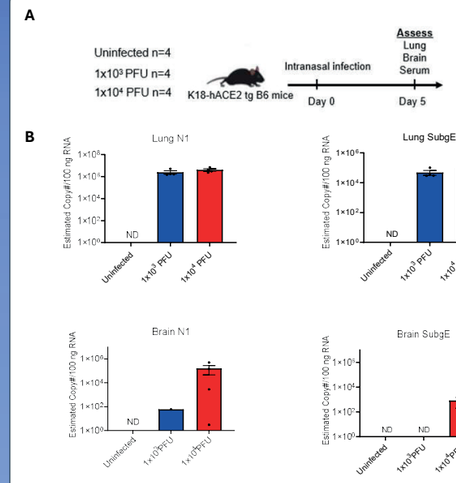


Human cytokines that are higher in the groups of ISARIC COVID-19 patients who had abnormal Glasgow Coma Scale Scores



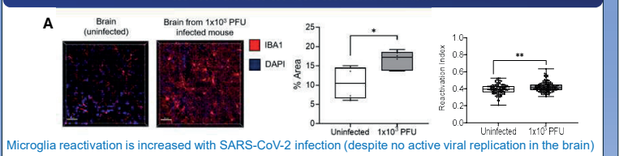
Deviation from Expected (DfE) Cognitron scores, split by diagnostic group in retrospective COVID-CNS cohort

Novel low-inoculum SARS-COV-2 infection mouse model

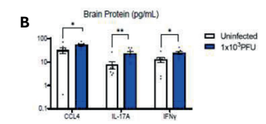


With low inoculum SC2 infection, no subgenomic E was present in brain tissue by RT-QPCR.

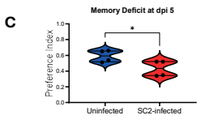
Immune activation in the absence of viral replication in the brain in the mouse model



Microglia reactivation is increased with SARS-CoV-2 infection (despite no active viral replication in the brain)



Increased cytokine expression in the brains of SARS-CoV-2 infected mice



Novel object recognition test preference index showing a memory deficit in SARS-CoV-2-infected mice at day 5 post-infection

CONCLUSIONS

- Two brain injury markers (NFL-L and UCH-L1) are elevated acutely following COVID-19 accompanied by an abnormal GCS
- Six cytokines/receptor (IL-1RA, IL-6, IL-12p40, HGF, M-CSF, and CCL2) are elevated acutely following COVID-19 accompanied by abnormal GCS
- We have established a mouse model of para-infectious SARS-CoV-2 effects on the brain– in the absence of viral replication in the brain, we observe increased cytokines (CCL4, IFN, IL-17A) and microglia reactivation (by increased Iba1 expression and morphology). This model will be useful for evaluating therapeutic strategies
- SARS-CoV-2 infection is accompanied by memory loss which is detectable in the mouse model by novel object recognition

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NIHR | National Institute for Health and Care Research

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Dr Crystal Sing Chiek Teoh

Department of Neurology, Queen's Medical Centre, Nottingham University Hospitals, UK

Clinical presentations and treatment outcomes of confirmed non-paraneoplastic seronegative and cell-surface antibody positive autoimmune encephalitis: experience from a tertiary neuroscience centre

Dr Crystal Sing Chiek Teoh is a motivated and aspired neurologist in training. Presently based in Queen's Medical Centre, Nottingham University Hospital as a neurology specialist registrar. She is interested in the fields of neuroimmunology especially in autoimmune encephalitis, multiple sclerosis/NMOSD/MOGAD, autoimmune neuropathies and neuromuscular junction disorders. Also interested in epilepsy, cognitive and vascular disorders. Active involvement in manuscript writing, research activities and systematic review. She has published several papers at peers reviewed international journals and presented poster in international congress. She participated in audit data collection, analysis, quality improvement projects and implementation of change. She underwent training in teaching and has experience in teaching junior doctors and medical students.

Clinical Presentations and Treatment Outcomes of Confirmed Non-paraneoplastic Seronegative and Cell-surface Antibody Positive Autoimmune Encephalitis: Experience from a Tertiary Neuroscience Centre

Crystal Sing Chiek Teoh[1], Afa Ibrahim[1], Beili Shao[1], Akram Hosseini[1], Bruno Gran[1], Radu Tanasescu[1]

[1]Department of Neurology, Queen's Medical Centre, Nottingham University Hospitals

INTRODUCTION

- Autoimmune encephalitis (AIE) is a heterogeneous group of inflammatory brain disorders characterized by immune-mediated brain inflammation.
- It includes both cell-surface antibody-positive AIE (SPAIE) and seronegative AIE (SNAIE), which can present with overlapping clinical features, such as cognitive dysfunction, seizures, and psychiatric symptoms.
- Data suggest that long-term outcomes between SPAIE and SNAIE may vary, with antibody-positive patients potentially experiencing more favourable responses to treatment.

AIM

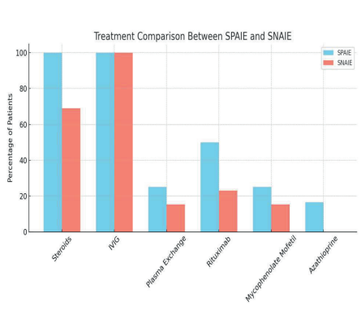
- This study aimed to evaluate and compare the clinical outcomes of patients with SPAIE and SNAIE at a tertiary neuroscience centre in the UK, with a focus on treatment strategies, relapse rates, and functional outcomes.
- As part of an audit, we concentrated on AIE cases treated with intravenous immunoglobulin (IVIG) at NUH NHS Trust between 2018 and 2023.

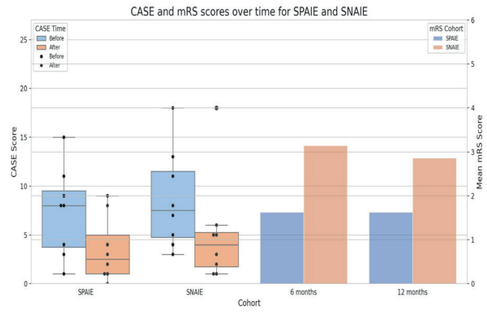
METHODS

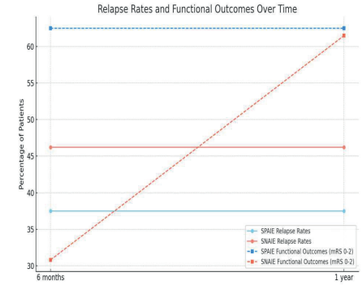
- We conducted a retrospective analysis of patients with confirmed AIE treated with IVIG between 2018 – 2023 (NUH 22-578C).
- All cases were curated against the 2016 AIE criteria. Data were collected on demographics, clinical presentations, investigation, treatment and outcomes. AIE mimics were excluded as per the Dalmau & Graus 2023 recommendations.
- Outcome measures included the Clinical Assessment Scale for AIE (CASE), modified Rankin Score (mRS), and the number of relapses.
- Univariate analyses were performed to identify differences between the groups.

RESULTS

- After individual case review, we identified 16 cases of confirmed AIE (8 SPAIE, 8 SNAIE) with a mean age of 39.75y (SD 19.8) and 32.62y (SD 24.9) respectively. Most patients were female (87.5% SPAIE, 76.9% SNAIE). No major differences were observed in clinical phenotypes, CSF , or MRI findings.
- The SPAIE group included 5 cases of NMDARAE, 1 CASPR2, 1 LGI-1 , and 1 TPO ab AIE. In addition to IVIG, first - line treatment included steroids (100% SPAIE vs 69% SNAIE) and plasma exchange (25% in SPAIE vs 15.4% in SNAIE). The use of second - line therapy with rituximab was double in SPAIE (50% vs 23.1%). Mycophenolate mofetil was used in 25% of SPAIE vs. 15.4% of SNAIE cases, and 16.5% of SPAIE patients used azathioprine. Antiseizure medication use was higher in the SNAIE (mean 1.1 in SPAIE vs 2.538 in SNAIE, p=0.014).
- Relapse rates at 1 year were 37.5% in SPAIE vs 46.2% in SNAIE. A favourable functional outcome (mRS 0-2) was achieved in 62.5% of SPAIE vs 30.8% SNAIE at 6 months, and 62.5% of SPAIE vs 61.5% SNAIE at 1 year. Excellent clinical outcomes (CASE score 0-4) were observed in 75 % of SPAIE vs 46% of SNAIE patients.







CONCLUSIONS

Acknowledging the limitation of small sample size, this retrospective study suggests more favourable outcomes in individuals with SPAIE compared to SNAIE. These findings underscore the importance of further research to better understand the disease mechanisms and develop biomarkers to better stratify these distinct patient populations. This will be key to optimizing treatment and enhancing long-term outcomes in AIE.

ACKNOWLEDGEMENT

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Protein biomarkers of neuroglial injury and inflammation in Herpes Simplex Virus Encephalitis

Dr Franklyn Nkongho Egbe is a Postdoctoral Research Associate at the Infection Neuroscience Laboratory, University of Liverpool. He is a Microbiologist with extensive experience in infectious disease research and clinical trials across Africa. His current research focuses on assessing inflammatory mediators and brain injury biomarkers in the blood and cerebrospinal fluid of patients with a wide array of infectious diseases that impact the nervous system like encephalitis, COVID-19 and epilepsy. His work is at the intersection of neuroscience and infectious diseases diagnostics, aimed at understanding how infections trigger neurological damage and how diagnostic assays can be improved for more effective patient outcomes. His research interests include infectious disease diagnostics, encephalitis, tuberculosis and COVID-19.

Protein biomarkers of neuroglial injury and inflammatory mediators in HSV encephalitis

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The Infection Neuroscience Lab

UNIVERSITY OF LIVERPOOL

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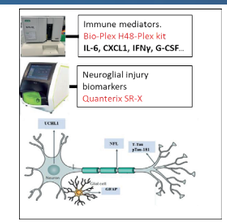
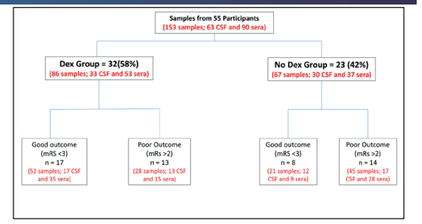
INTRODUCTION

Herpes Simplex Virus encephalitis (HSE) is brain inflammation with long-term neurodegenerative conditions caused by the Herpes Simplex Virus. Despite the effectiveness of antiviral therapy with acyclovir, fewer than 20% of patients achieve full recovery, with most experiencing enduring neurological disabilities[1]. There has been advocacy for using adjunctive immunomodulatory therapies to improve outcomes in HSV encephalitis by targeting the host inflammatory response, which is increasingly recognised as a critical component in HSE pathogenesis. Specifically, cytokines like the IL-1 family and its natural antagonist IL-1RA, and IL-10 have emerged as promising candidates for therapeutic modulation[2,3]. However, the complex mechanisms driving this inflammatory cascade and the full array of mediators involved remain poorly understood. We aimed to determine existing and novel potential adjunctive HSE therapy targets by exploring the relationship between host cytokine-mediated inflammatory mediators, neuroglial injury, and clinical outcomes in HSV encephalitis.

METHODOLOGY

We measured 48 inflammatory mediators and 4 neuroglial injury biomarkers in blood and cerebrospinal fluid (CSF) collected from 55 participants with Herpes Simplex Virus encephalitis enrolled in the DexEnceph clinical trial[4]; using the Bio-Rad Human 48 plex and Quanterix Neuro4plex bead-based digital ELISA assays respectively. Longitudinal sampling spanning the pre- and post-intervention period was done to determine the effect of Dexamethasone on the levels of these biomarkers. Clinical severity was assessed using the Glasgow Coma Scale score (GCS) at admission and the outcome measures were modified Rankin Score (mRS), and Liverpool Outcome Score; magnetic resonance imaging-based temporal lobe volumes. We determined the CSF HSV viral load using the RealStar HSV PCR Kit.





RESULTS

Mediators associated with clinical severity and outcome

CSF and serum IL-1RA; CSF CCL3; and serum IL-6 levels were associated with clinical severity and poor outcomes and had positive correlations with either MRI or GFAP, a marker of astrocytic damage. Fig. 1. The pro-inflammatory cytokine, CCL2 levels in serum and CSF were associated with clinical severity and the serum level showed a positive correlation with UCHL-1 (Fig. 2b) but was not associated with outcome.

On the other hand, the CSF level of CCL5 was associated with a mild coma and a good outcome. The anti-inflammatory chemokine, CXCL10 which can be produced by astrocytes in response to injury was associated with a mild disease but had no association with outcome.

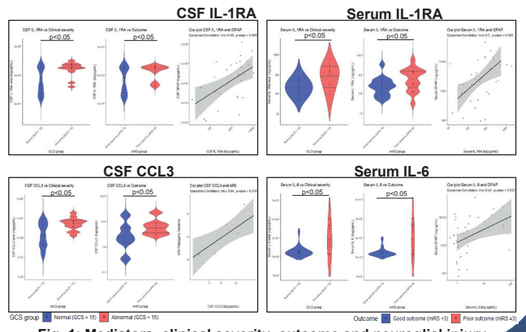


Fig. 1: Mediators, clinical severity, outcome and neuroglial injury

Mediators associated with MRI and neuroglial injury

In CSF, among mediators associated with either coma or poor disease outcomes, IL-1RA, IL-18, M.CSF, CCL3 and CXCL9 had positive correlations with raised neuroglial injury biomarkers ($p < 0.05$). Fig 2a

In serum, IL-1RA, IL-6, and M-CSF had positive correlations with raised levels of neuroglial injury biomarkers but no association with the MRI scans. Fig 2b

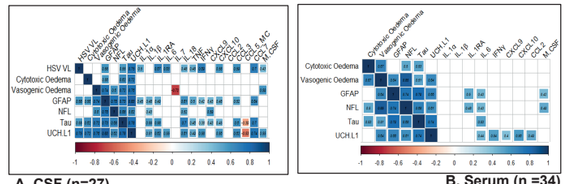


Fig. 2: Correlation matrix 0-9days post-randomisation

Change in mediators after treatment

Among 20 participants with at least 2 time point samples, we observed decreased IL-1α and M-CSF levels in dexamethasone treatment groups and increased levels in the placebo group. Fig 3

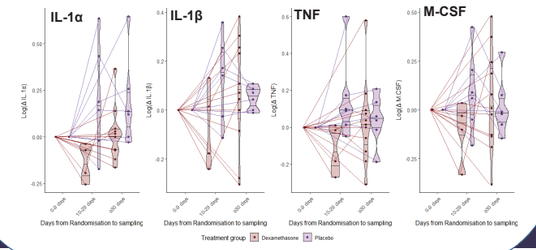


Fig. 3: Mediators fold change

Conclusions

CSF and serum levels of the IL-1 antagonist, IL-1RA were strongly associated with clinical severity, outcomes, brain swelling and neuroglial injury. The proinflammatory mediators, IL-18, IL-6, M-CSF, and CCL3 were associated with either clinical severity, poor outcomes or neuroglial injury. Predominantly proinflammatory cytokines and leucocyte chemokines are upregulated in HSV encephalitis. These markers could be potential targets for adjunctive therapy

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Use of bed-side cognitive tests in assessment of cognitive dysfuntion in autoimmune encephalitis

Mr Henrik Akre Thorup is a medical student doing a one-year undergraduate research study with Odense Autoimmune Encephalitis Research Group supervised by prof Morten Blaabjerg.

Use of bed-side cognitive tests in assessment of cognitive dysfuntion in autoimmune encephalitis

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BACKGROUND

Cognitive dysfunction is commen after recovery from autoimmune encephalitis (AE). Formal neuropsychological testing is the gold standard for evaluation of cognitive dysfunction, but is time comsuming and in many places not readily available. Simple bed-side tests including Mini-Mental State Examination (MMSE), Montreal Cognitive Assessment (MoCA) and Addenbrooke's Cognitive Examination (ACE) are easy and quick to administer. Their use in AE is, however, not investigated in detail. In this study, we correlated results from MMSE, MoCA and ACE to formal neuropsychological testing in a Danish cohort of AE patients.

METHODS

We recruited 78 participants from the Danish Autoimmune Encephalitis Study cohort (NMDAR n=36; LGI1 n=22; Caspr2 n=6; AMPA n=3, GAD65 n=3; GFAP n=4; GABAb n=1; GABAa n=1; DPPX n=1 and IgLON5 n=1). Formal neuropsycological testing included Trail making A and B, Reys Auditory Verbal Learning test, Reys Complex Figure Test, Wechsler Adult Intelligence Scale IV, D2-test, Delis-Kaplan Executive Function, Sentence Repetition Test and Symbol Digit Modalities Test. Linear regressions comparing averaged z-score from neuropsychological testing against the individual test scores from MMSE, MoCA and ACE were performed in the total cohort and different disease phenotypes (whole cohort, limbic encephalitis, NMDAR-AE and LGI1-AE subgroups).

RESULTS

Figure 1: Flowchart og patient inclusion and phenotypes

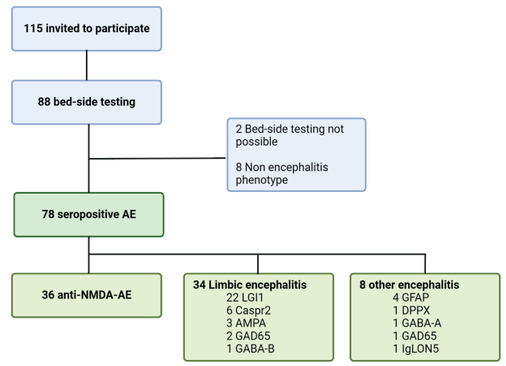
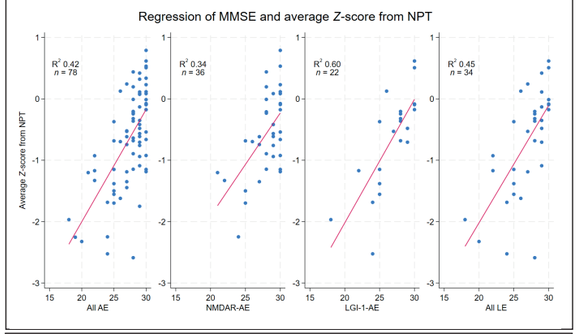
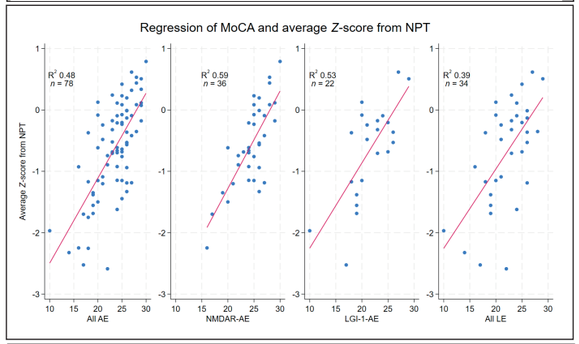


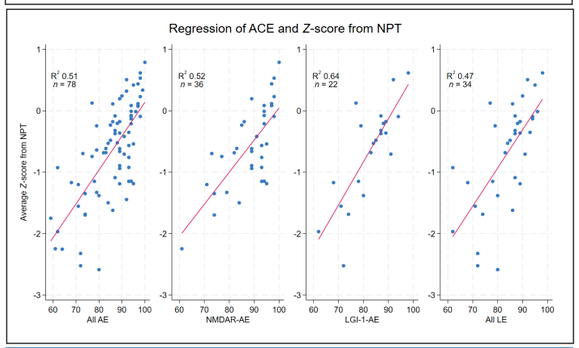
Figure 2: Demographics and phenotypes

	All patients n = 78	NMDAR-AE n = 36	Limbic encep. N = 34	LGI1-AE N = 22
Male sex n(%)	45(58)	15(42)	24(71)	12(55)
mRS IQR (median)	2(1)	1.5(0.5)	2(1)	2(2)
Age (y), IQR (median) Range	42(59) 18-90	26(32) 18-80	15(74) 30-90	10(76) 43-90
Time since diagnosis (y), IQR (median)	7.2(6.9)	5.3(6.4)	7.1(8.5)	5.8(10.3)

Regressions bed-side tests vs neuropsychological testing (NPT)







CONCLUSIONS

Bed-side cognitive tests (MMSE, MoCA and ACE) can be used in the follow-up and assessment of cognitive dysfunction in patients with AE as they correlate to results of formal neuropsychological testing. The choice of test should be based on disease phenotype. MoCA performs best in patients with NMDAR AE, whereas ACE performs best in patients with limbic encephalitis.

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Clinical description and outcomes of cryptogenic encephalitis in the intensive care unit

Dr Jackson Roberts is a recent graduate of Columbia University Vagelos College of Physicians and Surgeons and incoming neurology resident at Massachusetts General Brigham in Boston, MA. He has an interest in the epidemiology, diagnosis, and acute management of neuroinfectious diseases, particularly in the global context.

Characteristics and Acute Outcomes of Patients with Cryptogenic Encephalitis in the Neurological Intensive Care Unit

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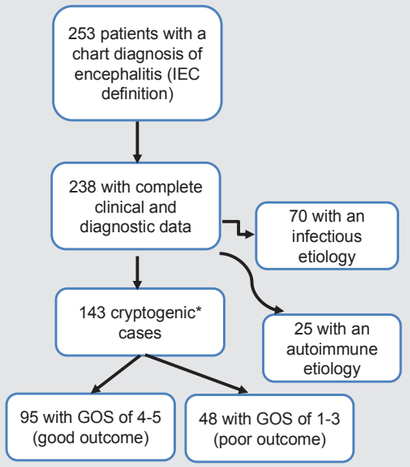
Background

- The majority of encephalitis cases are cryptogenic[1]
- Intensive care unit (ICU) encephalitis patients experience particularly poor outcomes, and little prognostic information is known for cryptogenic cases
- Better understanding this patient population will benefit family conversations and expectations in the ICU
- We sought to characterize cryptogenic ICU encephalitis cases and determine factors associated with poor prognosis

Participants and Methods



* Defined as no identified etiology by the time of discharge despite infectious and autoimmune workup

Methods

- Variables were selected for multivariate models by univariate comparison between good and poor outcome cases
- Multivariate logistic regression was performed between these variables and categorical Glasgow Outcome Scale at discharge (GOS)
- Multivariate linear regression was performed between these variables and continuous GOS at discharge

Clinical and Diagnostic Features

	Total (n=238)	Infectious (n=70)	Autoimmune (n=25)	Cryptogenic (n=143)	p-value
Age	41.55 (24.12)	45.19 (23.40)	26.56 (15.64)	42.49 (24.83)	0.003
Intubation	126 (52.94)	40 (57.14)	12 (48.00)	74 (51.75)	0.663
Status epilepticus	61 (25.63)	12 (17.14)	12 (48.00)	37 (25.87)	0.010
Multifocal lesion	94 (39.50)	25 (35.71)	14 (56.00)	55 (38.46)	0.189
Solitary lesion	10 (4.20)	5 (7.14)	0 (0)	5 (3.50)	0.249
Rhombencephalitis	14 (5.88)	4 (5.71)	2 (8.00)	8 (5.59)	0.892
Limbic Encephalitis	30 (12.61)	6 (8.57)	7 (28.00)	17 (11.89)	0.039
Meningeal Enhancement	42 (17.65)	23 (32.86)	5 (20.00)	14 (9.79)	<0.001
CSF profile obtained	160 (67.62)	47 (67.13)	17 (68.00)	96 (67.13)	0.227
CSF protein	167.38 (223)	284.7 (310)	54.43 (46.1)	128.0 (160)	<0.001
CSF glucose	76.81 (39.84)	65.82 (44.94)	71.80 (18.59)	81.89 (39.46)	0.032
CSF white blood cell count	532.7 (1969)	1154.8 (2990)	37.82 (73.16)	318.0 (1378)	0.0035
CSF percent lymphocytes	51.00 (36.47)	45.26 (37.29)	92.86 (7.47)	50.24 (35.26)	0.0018
Inpatient mortality	37 (15.54)	17 (24.29)	0 (0.00)	20 (13.99)	0.0058
GOS	3.72 (1.39)	3.60 (1.59)	3.76 (0.97)	3.77 (1.35)	0.812
Good outcome	151 (63.45)	42 (60.00)	14 (56.00)	95 (66.43)	0.471

Univariate Associations with Outcome

- Cryptogenic patients with a poor outcome were **older** (51.3 vs 37.9 years), had **longer ICU length of stay** (16.3 vs 7.1 days), **lower GCS** on admission (9.7 vs 11.6), were more often **immunocompromised** (58.3% vs 40.0%), more often had **active cancer** (12.5% vs 1.1%), and were more frequently **intubated** (72.9% vs 41.1%) than patients with a good outcome

Multivariate Associations with Outcome

A. Binary Glasgow Outcome Scale (i.e., Good vs Poor Outcome)

	Odds ratio (OR)	95% Confidence Interval	p-value
Age	0.974	0.957-0.992	0.005
ICU length of stay (days)	0.970	0.940-1.001	0.061
GCS on admission	1.035	0.915-1.171	0.582
Intubation	0.451	0.144-1.417	0.173
Active cancer	0.077	0.008-0.716	0.024
Meningeal enhancement	0.470	0.133-1.660	0.241

B. Continuous Glasgow Outcome Scale

	Unstandardized Coefficients		p-value
	Beta coefficient	Std. Error	
(Constant)	4.919	0.514	<0.001
Age (years)	-0.012	0.004	0.003
ICU length of stay (days)	-0.018	0.007	0.007
GCS on admission	-0.002	0.032	0.961
Intubation	-0.458	0.276	0.100
Meningeal enhancement	-0.770	0.332	0.022
Active cancer	-1.666	0.465	<0.001
Status epilepticus	-0.161	0.247	0.515

- Intubation and active cancer were also associated with a decreased likelihood of discharge to home or acute rehabilitation

Conclusions

- We identify that age and active cancer are associated with poor outcome, and meningeal enhancement and ICU length of stay are associated with GOS
- These factors may be used in prognostication with families in this heterogenous, unclassifiable population
- Similarities between cryptogenic and infectious cases suggest advanced diagnostic modalities (next generation metagenomic sequencing) may have utility in this population
- Future work should examine the post-discharge outcomes and possible diagnosis of etiology following hospitalization

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Dr João Moura

Department of Neurology, Hospital Santo António, Porto, Portugal

Characterization of EEG patterns in a cohort of patients with autoimmune encephalitis

Dr João Moura concluded his master's degree in medicine at ICBAS, University of Porto, in 2019. In 2021 he started his Neurology Residency at Hospital Santo António. His PhD project focuses on identifying novel anti-neural antibodies associated with autoimmune encephalitis from a national cohort of seronegative cases.

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Characterization of EEG patterns in a cohort of patients with autoimmune encephalitis

João Moura[1], Gonçalo Videira[1,2], Joana Lopes[1], Joel Freitas[2], Ester Coutinho[3,4,5], Raquel Samões[1], Ernestina Santos[1],

Introduction

Variable EEG features associated with autoimmune encephalitis (AIE) have been reported in the literature, the diagnostic /prognostic value of which remains unknown.

We aim to describe the EEG features from an institutional AIE cohort.

Methods

Retrospective review of the clinical and EEG features of AIE patients (according to Graus criteria) diagnosed between 2000 and 2023.

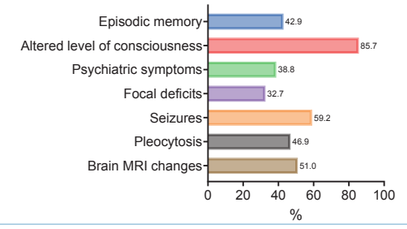
Results

Clinical Cohort

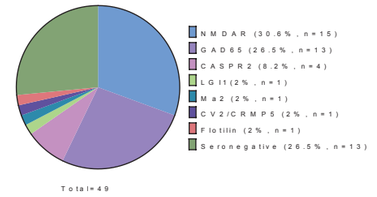
Demographics:

- 49 AIE cases
- 28♂
- Mean age at diagnosis 50.7 (±2.8) years
- Follow-up: mean 8.0 (±10.2) years

Clinical features:



Serogroups:



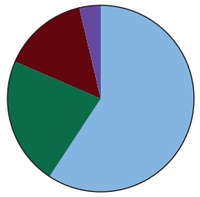
EEG Findings

EEG abnormalities in 40 (**81.6%**)

including 65% of patients without clinical seizures

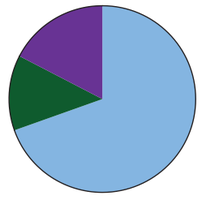
Slow activity 71.4%

Diffuse 20.4% / Focal 55.1%

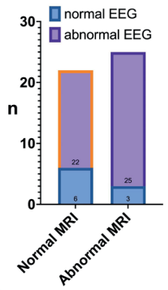


Temporal (59.3%, n=16)
Frontal (22.2%, n=6)
Fronto-temporal (14.8%, n=4)
Posterior (3.7%, n= 1)

Focal epileptiform activity 46.9%



Temporal (69.6%, n=16)
Frontal (13%, n=3)
Posterior (17.4%, n=4)



EEG abnormalities were equally common in patients with and without MRI abnormalities

No significant difference in the EEG features when comparing serogroups (including seronegative)

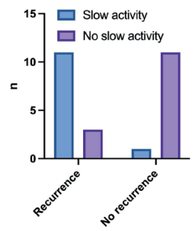
NCSE: 2 (4.1%) 1 NMDAr; 1 seronegative

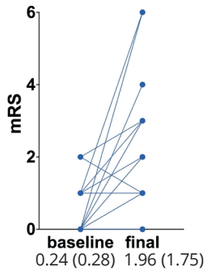
E*xtreme delta brush*: 3 (6.1%) 2 NMDAr; 1 seronegative

PLDs: 2 (4.1%) 1 CASPR2; 1 seronegative

Recurrence: 22.4%

Associated with **diffuse slow activity** in patients with intracellular abs (p=0.027)





Focal temporal activity associated with ↑ disability after 6.0 (3.0-8.0) years in patients with intracellular abs (p=0.042)

Conclusion

EEG abnormalities are common in AIE, even in cases with normal brain MRI or lacking clinical seizures.

Slow activity may be a prognostic factor, depending on the antibody type.

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Blackman G, et al. Quantitative EEG as a Prognostic Tool in Suspected Anti-N-Methyl-d-Aspartate Receptor Antibody Encephalitis. J Clin Neurophysiol. 2023
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Dr Jeroen Kerstens

Department of Neurology, Erasmus MC University Medical Center, Rotterdam, The Netherlands

Neuronal autoantibodies in atypical parkinsonism

Dr Jeroen Kerstens is a clinical neurologist with experience in autoimmune neurology and work as associated neurologist at the neuroimmunology unit of the Antwerp University Hospital, Belgium. In addition, He is a PhD candidate at the autoimmune encephalitis research group of the Erasmus MC University Medical Center in Rotterdam, the Netherlands. In 2016, he obtained his medical degree with great distinction at the University of Antwerp. Thereafter, he was trained in neurology in Antwerp and Rotterdam and completed his five-year residency in August 2021.

Neuronal autoantibodies in atypical parkinsonism

Jeroen Kerstens, j.kerstens@erasmusmc.nl

Jeroen Kerstens, MD[1], Agnita Boon, MD PhD[1], Elise Dopper, MD PhD[1], Laura Donker Kaat, MD PhD[2], Lieke Meeter, MD PhD[1], Yvette Crijnen, MD[1], Robin van Steenhoven, MD[1], Suzanne Franken, MSc[1], Mariska Nagtzaam, MSc[1], Harro Seelaar, MD PhD[1], John van Swieten, MD PhD[1] ,Juna de Vries, MD PhD[1], Peter Sillevis Smitt, MD PhD[1], Maarten Titulaer, MD PhD[1].

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Introduction

- Patients with antibody (Ab)-associated neurological disorders may lack typical characteristics of autoimmunity[1] and can be misdiagnosed as neurodegenerative diseases like progressive supranuclear palsy (PSP) or other forms of atypical parkinsonism.[2]
- One exemplary disease is anti-IgLON5, which can manifest chronic rather than subacute and ancillary investigations often do not show signs of inflammation.
- Correct diagnosis of patients with anti-IgLON5 disease or other Ab-associated syndromes may allow immunomodulatory treatment. This can provide considerable or even full recovery.

Main findings

- We identified 562 patients, of which 377 with available serum or CSF (Figure 1).
- 42/377 (11%) tested positive by IHC.
- Only 7/42 had Abs that could be confirmed with additional techniques and that were considered related to the patient's clinical syndrome (Table 1).
- We identified 23 Dutch anti-IgLON5 patients, nine of which had parkinsonistic or PSP-like features, but all of them also had atypical features that differentiated them from "classical" neurodegenerative parkinsonism.

Methodology

We retrospectively tested four patient cohorts (A-D) with suspected atypical parkinsonism:
- A: pts with suspicion of autoimmune parkinsonism (with or without other clinical features)
- B: pts referred to our tertiary movement disorders clinic between 2015 and 2024
- C: pts diagnosed with PSP between 2003 and 2014 according to NINDS-SPSP criteria
- D: deceased pts with clinical diagnosis of PSP but with discordant neuropathology

All pts were tested by immunohistochemistry (IHC); positive/questionable results were further evaluated with other techniques (cell-based assays (CBAs), live neurons, immunoblot).

Conclusions

- Routine testing of neuronal Abs in all atypical parkinsonism cases might not be warranted.
- Ab-associated disorders (anti-IgLON5 disease in particular) should remain in the differential diagnosis of atypical parkinsonism, especially in the presence of non-typical features.
- Positive IHC staining alone is insufficient for diagnosis of an Ab-associated disorder and could be explained by a non-specific immune reaction against epitopes released during a neurodegenerative disease process.

Results

Figure 1: Flowchart of all tested patients

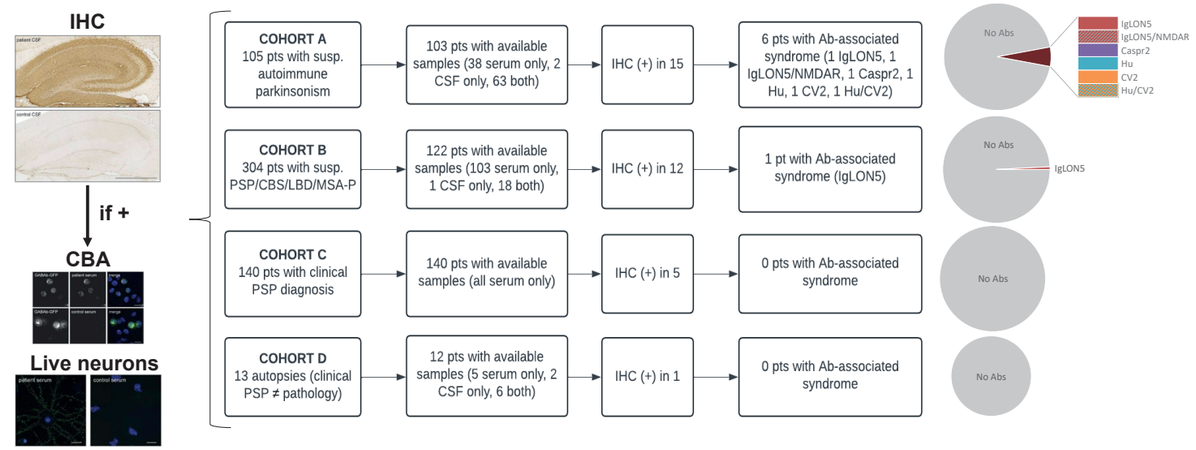

Table 1: Patients with Ab-associated parkinsonism

Cohort	Pt	Clinical syndrome	Ancillary tests	Tumor	Antibody testing	Final diagnosis	Treatment and response
A	M, 67y	PSP-like (parkinsonism, dementia, vocal cord paresis)	**Brain MRI**: microvascular white matter lesions **CSF**: wbc 2/µL, nl. protein, elevated IgG index	None	**Serum**: CBA anti-IgLON5 +, IHC + (neuropil) **CSF**: CBA anti-IgLON5 +, IHC + (neuropil)	Anti-IgLON5 disease	Partial improvement with immunotherapy
A	M, 48y	psychosis, isolated seizure, parkinsonism, autonomic dysfunction, severe mixed sleep apnea	**Brain MRI**: normal **CSF**: lymphocytic pleiocytosis (8/µL)	Seminoma	**Serum**: CBA anti-IgLON5 +, anti-NMDAR -, IHC + (neuropil), ELISA anti-GAD65 (113 IU/mL) **CSF**: CBA anti-IgLON5 +, anti-NMDAR +, IHC + (neuropil)	IgLON5/NMDAR-PNS	Improvement with immunotherapy and oncological treatment
A	M, 82y	limbic encephalitis plus (dementia, seizures, parkinsonism, autonomic symptoms)	**Brain MRI**: bilateral mesiotemporal T2 hyperintensities **CSF**: wbc 1/µL, nl. protein, no OCB	None	**Serum**: CBA anti-Caspr2 +, IHC + (neuropil), RIA anti-VGKC 343 pmol/L **CSF**: CBA anti-Caspr2 +, IHC + (neuropil)	Anti-Caspr2 encephalitis	Improvement with immunotherapy (IVMP, IVIg, oral prednisone, rituximab)
A	F, 77y	parkinsonism, myasthenia gravis	**Brain MRI**: small meningeoma (asymptomatic), posttraumatic sequelae **CSF**: not done	Thymoma	**Serum**: anti-nAChR + (26.2 nmol/L), anti-CV2 + (both IIF and immunoblot; titer 800) **CSF**: not done	CV2-PNS AChR+ myasthenia gravis	Partial improvement after oncological (resection) and symptomatic treatment (levodopa, pyridostigmine); no immunotherapy)
A	F, 73y	subacute parkinsonism	**Brain MRI**: three brain metastases (asymptomatic) **CSF**: unknown	SCLC	**Serum**: anti-Hu + (both IIF and immunoblot; titer 400) **CSF**: PNS immunoblot -	Hu-PNS	No improvement despite oncological (chemo- and radiotherapy) and symptomatic treatment (levodopa); no immunotherapy)
A	M, 72y	subacute parkinsonism	**Brain MRI**: bilateral T2 hyperintensities in basal ganglia **CSF**: lymphocytic pleiocytosis (17/µL)	SCLC	**Serum**: anti-Hu + (both IIF and immunoblot; titer 3200); anti-CV2 + (both IIF and immunoblot; titer 3200) **CSF**: anti-Hu + (both IIF and immunoblot); anti-CV2 + (both IIF and immunoblot)	Hu/CV2-PNS	Received oncological treatment (chemo- and radiotherapy); response unknown
B	M, 77y	MSA-P dd LBD (5y history of slowly progressive cognitive, bulbar, gait, sleep and autonomic symptoms)	**Brain MRI**: microvascular white matter lesions **CSF**: wbc 4/µL, nl. protein, elevated IgG index, CSF-specific OCB	None	**Serum**: CBA anti-IgLON5 +, IHC + (neuropil) **CSF**: CBA anti-IgLON5 +, IHC + (neuropil)	Anti-IgLON5 disease	Partial improvement with immunotherapy

Abbreviations: Abs: antibodies, CBA: cell-based assay, CBS: corticobasal syndrome, CSF: cerebrospinal fluid, ELISA: enzyme-linked immunosorbent assay, F: female, IHC: immunohistochemistry, IIF: indirect immunofluorescence, IVIg: intravenous immunoglobulins, IVMP: intravenous methylprednisolone, LBD: Lewy body disease, M: male, MSA-P: multisystem atrophy-parkinsonistic subtype, OCB: oligoclonal bands, PNS: paraneoplastic neurological syndrome, PSP: progressive supranuclear palsy, pts: patients, RIA: radioimmunoassay, SCLC: small-cell lung carcinoma, wbc: white blood cell

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ENCEPHALITIS CONFERENCE 2024

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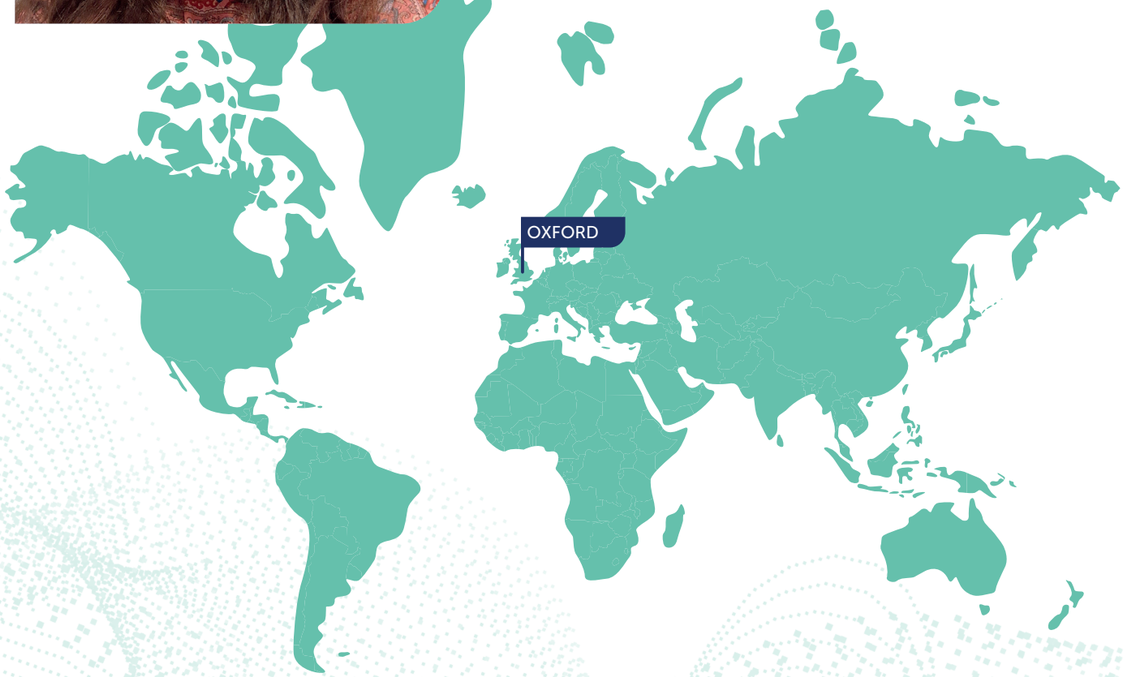
Phenotype, immunotherapy safety and outcomes in patients with autoimmune encephalitis following herpes simplex virus encephalitis: a systematic review.

Dr Jonathan Cleaver is currently a DPhil student working in the Oxford Autoimmune Neurology Group under Dr Adam Handel, Professor Sarosh Irani and Dr Lahiru Handunnetthi. His research area is exploring the immune mechanisms underpinning herpes simplex virus encephalitis and post-viral autoimmunity. They are currently using multi-modal biological approaches to understand how tolerance is breached in a subset of these patients together with developing biomarkers for those at risk of autoimmune encephalitis.

Miss Renetta Chungath is a third-year medical student at Lincoln College, University of Oxford, taking part in a Final Honours Scheme project focusing on Autoimmune Encephalitis with the Oxford Autoimmune Neurology Group under Dr Adam Handel.



Phenotype, immunotherapy safety and outcomes in patients with autoimmune encephalitis after herpes simplex virus encephalitis

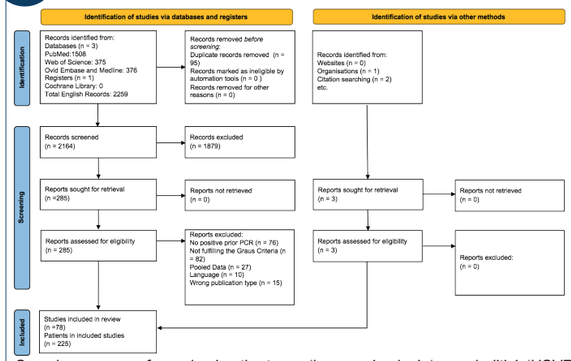
Jonathan Cleaver[1], Renetta Chungath[1], Amy Gimson[2], Thomas Johnson[3], Babak Soleimani[1,3], Lahiru Handunnetthi[1,3], Sarosh R Irani[1,4] Adam Handel[1]

Affiliations: 1) Nuffield Department of Clinical Neurosciences, Oxford, UK; 2) North Bristol NHS Trust, UK; 3) Centre for Human Genetics, Oxford, UK; 4) Departments of Neurology and Neurosciences, Mayo Clinic, US.

1 INTRODUCTION

- Herpes simplex virus encephalitis (HSVE) is the leading cause of sporadic encephalitis, with high mortality rate.
- Autoimmune encephalitis (AE) follows HSVE (HSVE-AE) in ~25% of cases, typically within 2 months.[1]
- HSVE-AE is predominantly associated with NMDAR antibodies.
- The known clinical phenotype is characterised by cognitive/behavioural changes, encephalopathy, seizures and movement disorders.
- It has been proposed that the clinical phenotype differs in young children <4 years versus older children and adults.[2]
- Factors influencing disability outcomes, as well as the outcomes themselves, are currently not well understood.
- We conducted a systematic review and meta-analysis to generate the largest dataset (n=225) of HSVE-AE patients to better understand the phenotype, immunotherapy safety and outcomes in this condition.
- We also compare this to a canonical NMDAR-antibody encephalitis database (not shown).[3]

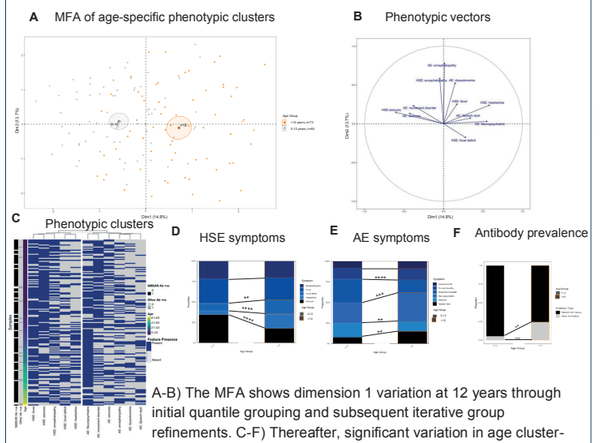
2 METHODS



Searches were performed using the terms 'herpes simplex', 'encephalitis', 'HSVE' and 'HSE'.

3 PHENOTYPIC DATA

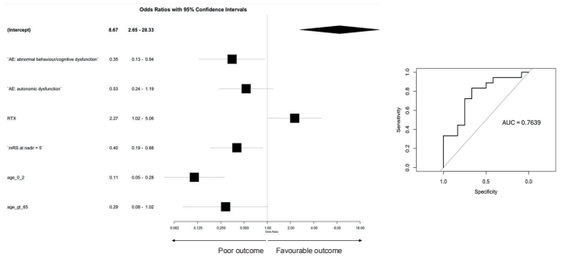
I. Multi-factor clustering analysis reveals age-specific phenotypic heterogeneity most marked at 12 years



A-B) The MFA shows dimension 1 variation at 12 years through initial quantile grouping and subsequent iterative group refinements. C-F) Thereafter, significant variation in age cluster-defined phenotypes emerge. MFA with headaches and speech dysfunction removed (not shown) maintain this age dichotomy.

4 REGRESSION ANALYSIS

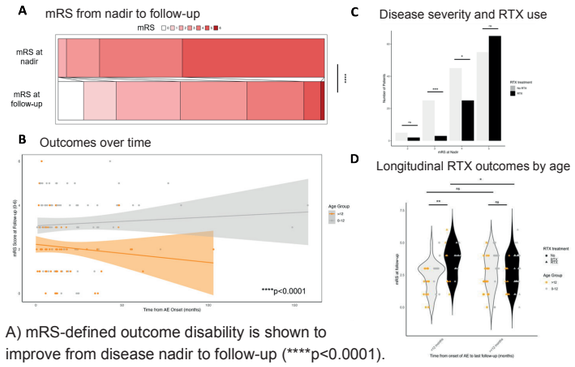
II. Multivariate logistic regression determines unique predictor variables for outcomes in HSVE-AE



Backward stepwise regression model with 10% K-nearest neighbour imputation isolated important predictor variables. The model was cross-validated with an 80-20 train-test split. A favourable outcome is defined as a follow-up modified Rankin score of 0-2 and poor outcome mRS >2. Non-imputed models and models including Armangue et al 2018[1] data returned similar results (not shown).

5 OUTCOMES

III. Overall longitudinal disability outcomes are favourable but with age- and treatment-specific influences



A) mRS-defined outcome disability is shown to improve from disease nadir to follow-up (****p<0.0001).
B) Individual follow-up time points show a clear longitudinal separation in outcomes between the younger, 0-12 years, and >12 years cohorts (****p<0.0001). C-D) RTX is predominantly administered at high nadir mRS which significantly reduces follow-up mRS at >12 months (*p=0.0154).

6 DISCUSSION

- We present the largest collection of HSVE-AE cases.
- Clinical phenotypes across ages are similar but heterogeneity is greatest at the age of 12 years.
- Cognitive/behavioural changes, mRS >4 at AE disease nadir and age 0-2 years are significantly associated with poor outcomes.
- Rituximab treatment is significantly associated with favourable outcomes.
- Outcomes significantly improve from disease nadir but outcomes are significantly worse in the 0-12-year cohort.
- Separate univariate analysis reveals marked changes in the clinical and paraclinical phenotype versus canonical NMDAR-antibody encephalitis.

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Dr Joseph Kuchling

Charité - Universitätsmedizin Berlin, Department of Neurology and Experimental Neurology, Berlin, Germany

7.0 Tesla MRI in NMDARE: Comparative Analysis of Supratentorial T2 Lesion Count and Hippocampal Subfield Volumes against Healthy Controls

Dr Joseph Kuchling is a post-doctoral researcher at the Cognitive Neurology Lab, Berlin (Prof. Carsten Finke) and a resident at the Department of Neurology and Experimental Neurology at Charité – Universitätsmedizin Berlin, Germany. After studying at Medical School Charité University Medicine Berlin and completing doctoral studies on ultrahigh-¬field MRI in multiple sclerosis, Joseph has been working in the field of advanced neuroimaging including diffusion tensor imaging and 7 Tesla MRI in neuroinflammatory diseases.

7.0 Tesla MRI in NMDARE:

Comparative Analysis of Supratentorial T2 Lesion Count and Hippocampal Subfield Volumes against Healthy Controls

Joseph Kuchling, MD; Josephine Heine, PhD, Friedemann Paul, MD, Thoralf Niendorf, PhD, Carsten Finke, MD

Department of Clinical and Experimental Neurology, Charité-Universitätsmedizin Berlin

COGNITIVE NEUROLOGY LAB
Charité - Universitätsmedizin Berlin

CHARITÉ UNIVERSITÄTSMEDIZIN BERLIN

Introduction

Patients with anti-N-methyl-D-aspartate receptor encephalitis (NMDARE) often show T2-hyperintense lesions and hippocampal atrophy, correlating with memory performance and disease severity.

7.0 Tesla (7T) MRI provides increased spatial resolution and higher tissue contrast compared to 3T MRI.

Finke et al. 2013, Ann Neurol; Le Ster et al. 2022, Magn Reson Med; Hechler A, Kuchling J, et al. J Neurol 2024

Objectives and Methods

I. Supratentorial T2(*) lesion count at 7 T and 3 T
- *2D T2*w FLASH (0.5×0.5×2.0 mm³)*
- *Horos medical image viewer (version 4.0.0)*

II. Analysis of hippocampal subfield volumes in NMDARE and HC at 7 T
- *T1 MPRAGE (resolution: 1x1x1 mm³)*
- *Freesurfer (version 6.0)*

III. Comparison of hippocampal subfield volumes in NMDARE at 7 T and 3 T

Cohort

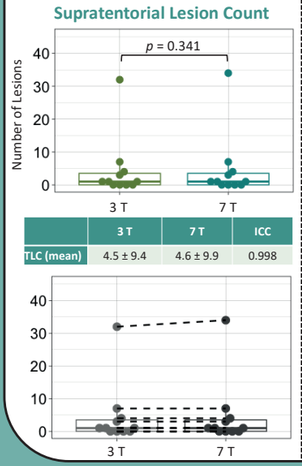
11 NMDARE patients were investigated at 7 T and 3 T

11 HC were investigated at 7 T

	NMDARE	HC	p
n	11	11	
Sex (f; %)	10 (91 %)	8 (73 %)	0.580
Age (mean; SD)	35.9 (9.8)	34.0 (12.7)	0.683

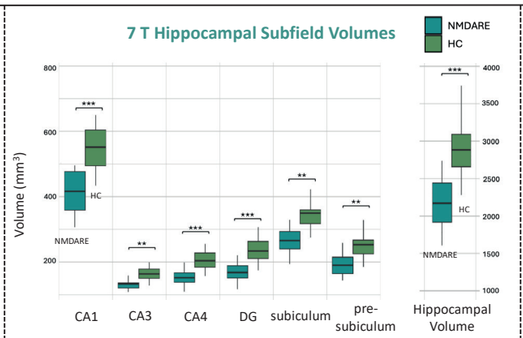
Results

Supratentorial Lesion Count



	3 T	7 T	ICC
TLC (mean)	4.5 ± 9.4	4.6 ± 9.9	0.998

7 T Hippocampal Subfield Volumes

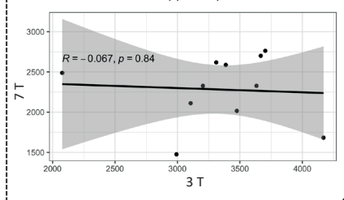


	NMDARE	HC	p-value (t-test)	AUC (ROC)
Hippocampus	2150 ± 360 mm³	2871 ± 434 mm³	<0,001*	0.843
CA1	415 ± 67 mm³	549 ± 77 mm³	<0,001*	0.818
CA3	117 ± 27 mm³	158 ± 23 mm³	<0,001*	0.826
CA4	146 ± 27 mm³	199 ± 32 mm³	<0,001*	0.818
Dentate Gyrus	174 ± 31 mm³	237 ± 39 mm³	<0,001*	0.826
Subiculum	263 ± 46 mm³	346 ± 63 mm³	<0,001*	0.818
Presubiculum	190 ± 37 mm³	253 ± 56 mm³	<0,001*	0.785

Comparison of 7 T and 3 T

	ICC	p-value (ICC)	Pearson correlation	p-value (Pearson)
Hippocampus	-0.07	0.580	-0.07	0.844
CA1	-0.27	0.802	-0.29	0.393
CA3	0.00	0.504	0.00	0.993
CA4	-0.17	0.704	-0.18	0.605
Dentate Gyrus	-0.20	0.730	-0.20	0.547
Subiculum	-0.09	0.610	-0.09	0.788
Presubiculum	-0.41	0.906	-0.41	0.213

Bilateral Hippocampal Volume



Conclusion

7T MRI facilitates the detailed volumetric measurement of hippocampal subfields in NMDARE patients, showing significantly lower volumes in NMDARE compared to healthy controls.

7T and 3T hippocampal (subfield) volumes showed poor agreement and no advantage of 7T over 3T was found for detecting supratentorial T2 lesions.

More research is needed to assess the full benefits of 7T MRI in NMDARE including correlation analyses between 7 T MRI imaging and clinical outcome.

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ENCEPHALITIS CONFERENCE 2024

Dr Kate Halsby

Pfizer Ltd, Surrey, UK

Need for a Categorisation System of the Clinical Manifestations of Tick-borne Encephalitis Virus Infection: A Delphi Panel

Dr Kate Halsby is an epidemiologist working in Pfizer's Global Vaccines and Anti-infectives medical team, with a focus on tick-borne encephalitis and Lyme disease. Before moving into industry, she worked on zoonotic infections and Legionnaires' disease at Public Health England.

Need for a Categorisation System of the Clinical Manifestations of Tick-borne Encephalitis Virus Infection: A Delphi Panel

Presenter: Kate Halsby
Email: kate.halsby@pfizer.com

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BACKGROUND

Tick-borne encephalitis (TBE) is a disease characterized by inflammation of the central nervous system (CNS) resulting from infection with the TBE virus (TBEV).[1]

Disease presentation for TBE is commonly categorized as: Meningitis ("mild"), encephalitis ("moderate"), and encephalomyelitis ("severe"). However,

1. Meningitis can be severe
2. Many other categorization systems are used (**Figure 1**)

This heterogeneity in classification creates issues for comparison of data between studies, and a lack of consistency in descriptions of severity for TBE.

METHODS

A 10-member panel of experts on TBE including clinicians (including pediatricians), epidemiologists and health scientists from 8 European countries was convened to:

- Determine the need for a clinical categorization system for TBE
- Outline requirements for such a system

Delphi methodology[6] has a structured & controlled feedback process that encourages panelists to reassess their initial judgements. It also emphasizes anonymity of panelists in order to avoid direct confrontation and ensure response confidentiality.

Experts responded to two rounds of surveys and attended a virtual consensus-building exercise, receiving feedback about the previous round prior to each subsequent round (**Figure 2**).

Figure 2: Delphi Methodology as Applied to Project

Initial Questionnaire	Gather information and feedback from panelists
Second Questionnaire	Present results of questionnaire 1 to panelists and pose follow-up questions to elicit deeper feedback
Consensus-building Exercise	Identify areas of agreement and discuss areas of disagreement to reach accord

RESULTS

The outputs of the two questionnaires is shown in **Figure 3**. Four consensus statements were agreed:

- It is important to have a general standard categorization system for patients infected with TBEV that optimally reflects both the clinical presentation and severity of illness of such patients
- There is no existing classification system that effectively captures both the clinical presentation and severity of TBEV infection
- A two-part system that separately captures the acute and follow-up (outcome) phases of TBEV infection is needed
- There needs to be a clinically oriented classification system with an overall score with ranges depicting severity of TBEV infection

The panel further aligned on the considerations for a TBE classification system (**Figure 4**).

AREAS FOR FURTHER DISCUSSION

i) The ideal timepoint for follow-up. There was greatest agreement on the 12-month mark, but a desire to capture follow-up at multiple timepoints.

> *"Minimum 12 months, if needed longer."*

> *"Has to be multiple timepoints as it will differ at 6 months to 3 years. You can't say the outcome at 6 months is the same as outcomes at 3 years."*

ii) Whether children and adults should have different systems or timepoints.

- Lack of meaningful cut-off age for children, e.g., a 17-year-old may manifest the same as an adult
- Difficult to detect some symptoms in younger children (e.g., neck stiffness)

> *"Definitely two separate systems. Children are not small adults. Small children are still developing."*

> *"Symptoms of tick-borne encephalitis are non-specific and the same in all age groups, so it is not necessary to classify the severity of the disease separately for children."*

> *"Yes, I think paediatric and adult populations often present differently."*

> *"We need to be able to compare the data. And at what age would the line be drawn? Although the disease severity generally differs between children and adults, it makes more sense to use the same system."*

Figure 1: Varying Classification of TBE CNS Infection in the Literature

Santonja et al., 2022[2]	
Mild	Febrile illness and meningitis
Severe	Meningoencephalitis, encephalomyelitis, and radiculitis

Bogovic et al., 2014[3]	
Mild	0–8 points on symptom score scale
Moderate	9–22 points on symptom score scale
Severe	>22 points on symptom score scale

Hansson et al., 2020[4]	
Mild	Signs of meningitis and normal electroencephalography when applicable
Moderate	Mild signs of altered consciousness with diffuse symptoms such as slow cerebration, confusion, and/or focal neurological signs such as ataxia, tremor, and dysphasia
Severe	Severe signs of altered consciousness and/or multifocal neurological symptoms

Pichler et al., 2017[5]	
Mild	Meningeal symptoms including fever, headache, rigidity of the neck, and nausea
Moderate	Monofocal deficits of the CNS and/or moderate dysfunction of the brain including tremor, ataxia, dysphagia, single cranial nerve affection and moderate decline of vigilance
Severe	Multifocal deficits of the CNS and/or severe brain dysfunction including seizures, central paralysis, multifocal cranial nerve deficits, and affection of the spinal cord

Figure 3: Questionnaire Outputs

1st Questionnaire	2nd Questionnaire
Purpose	**Purpose**
Broad, covering: i) Existing categorization systems for TBE ii) Important characteristics of TBE	Targeted, exploring the appropriateness of different categorization systems
Highlights	**Highlights**

• **It is important to have a standard categorisation system that reflects both clinical presentation and severity of illness (100% agreement)**
  • 40% agreement: a clinical categorisation system exists
  • 80% agreement: a system to categorise severity exists
  • 30% agreement: that a system to do both exists
• TBE has both acute and long-term impacts
• Characteristics to include in a categorization system, (≥90% agreement):
  • Meningitis / encephalitis / myelitis / radiculitis
  • Impact on daily life
  • Sequelae
	• A two-part system (acute and long-term) is appropriate (100% agreement)
	• Acute: the following factors may be considered (≥90% agreement):
	  • Alteration of consciousness
	  • Respiratory insufficiency
	  • Limb paresis
	  • Fever (with/without CNS involvement)
	• Follow-up: the following factors may be considered (≥90% agreement):
	  • Clinical, including neurological, factors
	  • Quality-of-life indicators
	  • Disability and functional recovery

Figure 4: Considerations for a TBE Classification System

The Overall System Should…
…optimally reflect both clinical presentation and severity of illness
…not conflate mild, moderate, and severe with meningitis, encephalitis, and encephalomyelitis
…be easy to implement, simple, and practical
…be standardised for comparability purposes
…be one system that can differentiate children vs. adults while recognizing that differences exist (e.g., fever must be interpreted carefully in children, adult, and older patients)
…be properly developed and endorsed by professional societies

The Acute Categorization System Should…	The Follow-up Categorization System Should…
…be useable by any treating clinician, regardless of specialty	…be useable by all types of TBE providers and clinical researchers
…include clinical manifestations, neurological symptoms, and respiratory function.	…focus on functional outcomes and activities of daily living and may include impact on quality of life
…include CSF (should) and MRI findings (could), if clinically required	…include remission and progression factors
…include alteration of consciousness, neurological dysfunction, respiratory insufficiency, and fever	…require a minimum of 12 months of follow-up (or longer depending on clinical manifestation, severity, and sequelae)
…be useable at bedside	…be able to be captured using elements from the patients' medical record (as best practice)

CONCLUSIONS

There is no existing classification system that effectively captures both the clinical presentation and severity of TBEV infection.

The needed system should:

- Separately capture the acute and follow-up phases of symptomatic TBEV infection
- Be clinically oriented with an overall score depicting severity of symptomatic TBEV infection

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ENCEPHALITIS CONFERENCE 2024

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The role of microglia in pneumococcal meningoencephalitis

Miss Kristine Farmen has a Masters degree in molecular medicine focusing on the role of peripheral immune cells in Parkinson's disease. Currently doing a PhD in neuroscience with the focus on understanding the biological mechanism of neuronal cell damage during brain infections and developing new therapeutic targets against pneumococcal meningoencephalitis.

The role of microglia in pneumococcal meningoencephalitis

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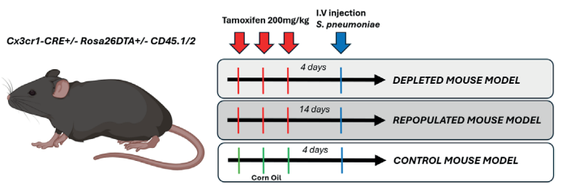
Background

During the pathogenesis of pneumococcal meningoencephalitis, *Streptococcus pneumoniae* cause direct cellular damage in the brain by the release of virulence factors; however, most of the cellular damage is due to the inflammatory response in the brain, caused by an activation of microglia, and the infiltration of peripheral immune cells, such as neutrophils and monocytes. **The aim** of this study was to describe the impact of the presence of microglia on the *S. pneumoniae* load in the brain, to better understand if microglia play a pivotal or deleterious role in the removal of the bacteria; as well as how they contribute to the inflammatory milieu and peripheral immune cell infiltration.

Experimental setup and results



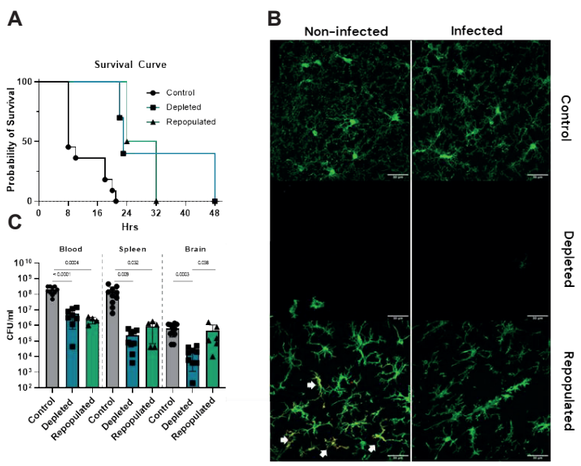


Figure 1. Alteration in microglia dynamics and number (B) increase survival rate (A) and lowers bacterial load in the periphery and in the brain (C).

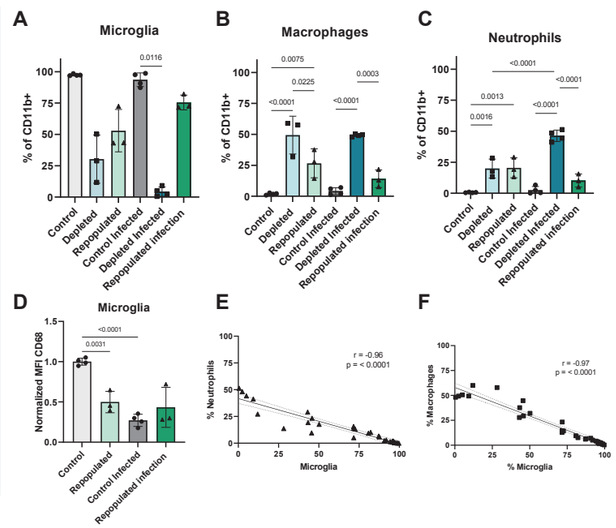


Figure 2. Depletion of microglia (A) results in increased macrophage (B) and neutrophils levels in the brain (C). During infection, the phagocytic marker CD68 is reduced (D). The levels of microglia correlates with the levels of macrophages (E) and neutrophils (F) in the brain.

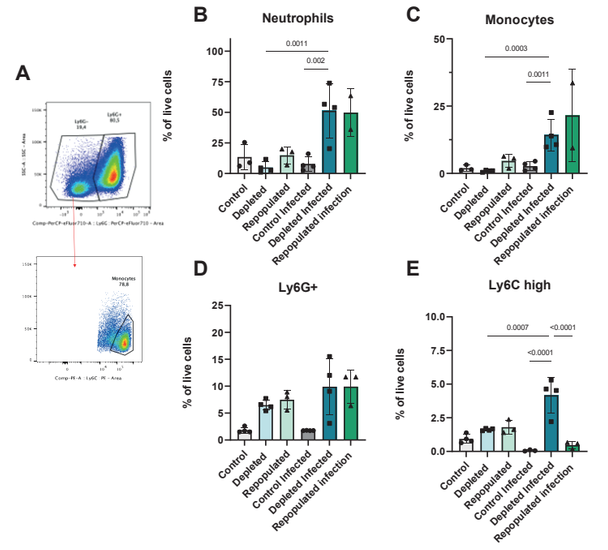


Figure 3 Analysis of the peripheral innate immune system, gating strategy (3A), showed increase in blood neutrophils B) and monocytes (C) and increase in spleen Ly6G+ D) and Ly6C E) high in depleted and repopulated mice during infection.

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ENCEPHALITIS CONFERENCE 2024

Mr Leonardo Di Cosmo

Oxford Autoimmune Neurology Group, Nuffield Department of Clinical Neurosciences, University of Oxford, UK

A systematic review of the clinical features associated with seronegative autoimmune encephalitis.

Mr Leonardo Di Cosmo is currently a Medical Student at Humanitas University, Milan, where he is actively involved within projects and roles involving the neuroscientific field. He is currently the Seminar Director of the Humanitas Neurological Society and leads a research group affiliated with the Mission Brain foundation. Prior to entering Medical School, Leonardo Di Cosmo founded a non-profit in the Philippines that worked alongside the Philippine General Hospital to provide prosthetics and bone marrow transplants for paediatric patients in the oncology ward. He is committed to exploring the field of research, to date having interned at multiple cancer-immunology labs at Humanitas University. Though only at the start of his medical career, he is a Visiting Researcher at the Nuffield Neurological Clinic at Oxford University, currently leading a first-of-its-kind systematic review exploring the clinical presentation of seronegative encephalitis. Furthermore, despite his relatively short time in the field of encephalitis, Leonardo is dedicated to continuing cultivating his knowledge and practice in pursuit of making meaningful contributions to the field.

CLINICAL CHARACTERISTICS AND OUTCOMES IN SERONEGATIVE AUTOIMMUNE ENCEPHALITIS

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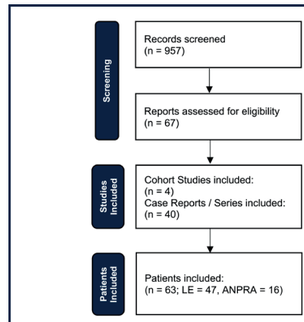
BACKGROUND

- **Clinical Significance**: The absence of identifiable autoantibodies complicates the **diagnosis**, **prognosis**, and **management** of patients with autoimmune encephalitis (AE). The seronegative group comprises **33–50%** of all autoimmune encephalitis cases[1]. While the Graus criteria[2] provide a foundation for diagnosis, a more tailored guideline specific to seronegative cases would be impactful in both research and clinical practice.
- Therefore, our **systematic review and meta-analysis** aims to evaluate the **clinical characteristics**, **immunotherapeutic responses**, and **outcomes** in patients with seronegative autoimmune encephalitis (SN-AE), including those diagnosed with seronegative limbic encephalitis (**LE**) and antibody-negative probable autoimmune encephalitis (**ANPRA**).

METHODS

- **Systematic Literature Search** of case reports, case series, cohort studies, and quasi-experimental studies published between **2014-2024**, focusing on adults diagnosed with seronegative autoimmune encephalitis based on Graus criteria[2].
- **Screening and Data Extraction** were conducted by two independent reviewers, extracting data on clinical profiles and outcomes.
- **Additional Clinical Data** was obtained from patients treated at John Radcliffe Hospital in Oxford to supplement the literature findings.
- **Statistical Analysis** was performed using R (RStudio, version 4.4.1). Descriptive statistics including means and medians were calculated and comparative analyses such as chi-squared tests, t-tests, and Mann-Whitney U tests were conducted where appropriate. A linear regression model was also generated to explore associations between clinical variables and outcomes.

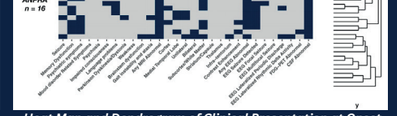


SERONEGATIVE AUTOIMMUNE ENCEPHALITIS CLINICAL PRESENTATION AND OUTCOMES

	Total (n = 63)	LE (n = 47)	ANPRA (n = 16)	p-values
Age (mean, median)	53.2, 59.0	51.8, 55.0	57.4, 61.5	0.129
Male sex (%)	54.0% (34)	53.2% (25)	56.2% (9)	1.000
Initial CASE scores (mean, median, n)	8.0, 7.0 (48)	7.5, 6.5 (32)	8.9, 8.5 (16)	0.118
Initial mRS scores (mean, median, n)	3.6, 3.0 (59)	3.5, 3.0 (45)	3.8, 4.0 (14)	0.250
Seizure (%)	50.8% (32)	51.1% (24)	50.0% (8)	1.000
Memory dysfunction (%)	85.7% (54)	89.4% (42)	75.0% (12)	0.320
Psychiatric symptoms (%)	58.7% (37)	59.6% (28)	56.2% (9)	1.000
Mood-disorder Symptoms (%)	33.3% (21)	40.4% (19)	12.5% (2)	0.082
Psychosis (%)	22.2% (14)	23.4% (11)	18.8% (3)	1.000
Impaired consciousness (%)	33.3% (21)	29.8% (14)	43.8% (7)	0.810
Language problem (%)	27.0% (17)	19.1% (9)	50.0% (8)	0.067
Dyskinesia/dystonia (%)	17.5% (11)	14.9% (7)	25.0% (4)	0.583
Gait instability and ataxia (%)	14.3% (9)	10.6% (5)	25.0% (4)	0.357
Brainstem dysfunction (%)	7.9% (5)	8.5% (4)	6.2% (1)	1.000
Weakness (%)	6.3% (4)	8.5% (4)	0.0% (0)	0.657
First Line Immunotherapy (%)	95.2% (60)	93.6% (44)	100.0% (16)	0.722
IVIG (%)	38.1% (24)	42.6% (20)	25.0% (4)	0.448
Corticosteroids (%)	87.3% (55)	83.0% (39)	100.0% (16)	0.231
Plasmapheresis (%)	17.5% (11)	14.9% (7)	25.0% (4)	1.000
Second Line Immunotherapy (%)	25.4% (16)	25.5% (12)	25.0% (4)	1.000
Rituximab (%)	17.5% (11)	14.9% (7)	25.0% (4)	0.580
Cyclophosphamide (%)	4.8% (3)	0.0% (0)	6.2% (1)	0.498
FLIT CASE Scores (mean, median, n)	4.4, 4.0 (46)	4.0, 4.0 (31)	5.1, 4.0 (15)	0.367
FLIT mRS Scores (mean, median, n)	2.6, 2.0 (61)	2.6, 2.0 (46)	2.5, 2.5 (15)	0.993
SLIT CASE Scores (mean, median, n)	2.3, 1.5 (12)	1.7, 1.0 (9)	4.3, 4.0 (3)	0.772
SLIT mRS Scores (mean, median, n)	2.3, 2.0 (14)	2.1, 2.0 (11)	3.0, 3.0 (3)	0.574

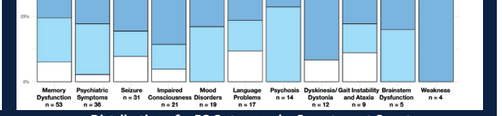
Clinical and Paraclinical Characteristics

This table presents a breakdown of the demographic characteristics of patients included in the review. It showcases the distribution of key variables encompassing clinical presentation between the two patient populations of seronegative limbic encephalitis (LE) and probable AE (ANPRA). Additionally, it presents descriptive analyses of post-immunotherapeutic patient outcomes, and respective uncorrected p values.



Heat Map and Dendrogram of Clinical Presentation at Onset

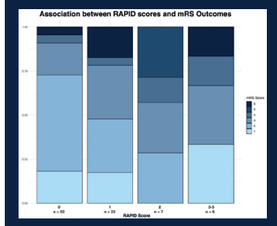
This heat map displays the constellation of clinical symptoms and diagnostic findings in our patient sample at the time of initial presentation, including symptom profiles, MRI, EEG, FDG-PET, and CSF results. The dendrogram reveals patterns in symptomatology that distinguish seronegative limbic encephalitis (LE) from probable AE (ANPRA), offering insights into symptom groupings that may aid in diagnostic modelling.

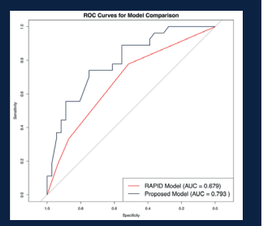


Distribution of mRS Outcomes by Symptom at Onset

This bar chart shows the distribution of mRS outcomes post-first line immunotherapy (FLIT) across the patients, relative to symptom presence at onset. This figure highlights which initial symptoms are linked to more favorable or severe disability outcomes. However, frequencies are unadjusted for the number of symptoms each patient experienced. Each variable underwent linear regression to normalize for patient age and sex.

Predictive Models of Clinical Outcomes





RAPID

Predictor of Poor Outcome (mRS≥3)	Odds Ratio (95% CI)	p-value
Refractory Status Epilepticus (RSE)	1.81 (0.28 - 14.62)	0.533
Age of Onset ≥ 60 years	2.65 (0.93 - 7.89)	0.073
Probable AE (ANPRA) Subtype	2.19 (0.63 - 8.23)	0.224
Infra-tentorium Involvement	0.62 (0.07 - 5.89)	0.663
Delay of Immunotherapy ≥ 1 month	4.38 (0.53 - 94.96)	0.223

Proposed Model

Predictor of Poor Outcome (mRS≥3)	Odds Ratio (95% CI)	p-value
Age of Onset ≥ 60 years	4.25 (1.19 - 17.16)	**0.031**
Severe Disability at Onset (mRS ≥ 4)	4.27 (1.11 - 19.23)	**0.044**
Psychosis	0.13 (0.02 - 0.71)	**0.028**
Mood-Disorder	0.14 (0.02 - 0.62)	**0.018**
Gait Instability and Ataxia	2.64 (0.43 - 17.45)	0.294
Brainstem Dysfunction	4.65 (0.53 - 51.42)	0.172
Symptom Development Prior to Admission (Days)	1.01 (1.00 - 1.02)	0.159
Corticosteroid Use	0.08 (0.01 - 0.68)	**0.029**

- A binomial generalized linear model (glm) analysis was conducted to model the relationship between predictors and the clinical outcome variable, representing the patient's clinical status after first-line immunotherapy (FLIT). The binary outcome measured the patient's clinical status after first-line immunotherapy (FLIT), with scores ≥3 indicating poor outcomes and <3 indicating favorable outcomes..
- The RAPID model, proposed by Lee et al.[3] as a predictor of patient outcomes, includes five variables associated with poorer outcomes. This model was applied to our cohort and compared to another developed from our patient data. This enabled us to identify discrepancies in the predictors of clinical outcome between the two models.

RESULTS

- A **dichotomy in clinical presentation** between the seronegative groups was observed: the **ANPRA** group exhibited a **higher proportion of language deficits**, while the **LE** subgroup showed a **higher proportion of mood disorder-related symptoms**. Although these differences did not reach statistical significance, they were **marginally significant**. Other discrepancies were noted, but none were statistically significant possibly due to imbalanced sample sizes.
- The distribution of mRS outcomes by symptom revealed patients with **brainstem dysfunction**, **gait instability and ataxia**, and **impaired consciousness** were associated with **higher mRS scores** , with a significant proportion having mRS ≥ 4. **Mood disorders** and **psychosis** were associated with more favorable mRS outcomes, having the greatest proportion of mRS 1 - 2. This result may derive from the mRS scale's emphasis on physical disability, which may underrepresent the impact of psychological symptoms such as mood disorder and psychosis. **Memory dysfunction**, **psychiatric symptoms**, and **seizures** show a **wider distribution** across mRS scores, indicating **variability in patient outcomes**. This may be because these symptoms were the most frequently represented, facilitating a more normalized distribution.
- The proportion of **poor mRS outcomes** across **RAPID scores** shows a moderate distribution. Patients with a RAPID score of **0 mostly had favorable mRS outcomes**, while those with higher RAPID scores (especially 2 and 3-5) exhibited **more severe disability or death (mRS ≥ 4)**. However, there are **notable exceptions to this distribution**. RAPID = 1 had the **greatest proportion of deaths (mRS = 6)**, and RAPID = 2 had the **highest proportion of mRS ≥ 5**. However, it is important to acknowledge the significant heterogeneity between the size of each group, with most presenting with RAPID 0 - 1. This may be due to the **gaps in comprehensive reporting** in case studies, as well as the **limited amount of ANPRA cases** in our population, which itself is a variable considered in the RAPID score.
- The **RAPID model** showed moderate effectiveness in predicting patient outcomes (**AUC = 0.679**), and our proposed model demonstrated a **higher predictive accuracy** in this specific cohort (**AUC = 0.793**). This result is expected given our model was developed and tested on the same dataset. It is also important to note that **RAPID scores** were based on mRS outcomes after **2-year follow-ups**, whereas the follow-up time in our cohort was **heterogeneous** and often **poorly reported**. Additionally, our model focused on **post-FLIT patients**, while the RAPID model incorporated both **post-FLIT and second-line immunotherapy (SLIT) patients**. Furthermore, in clinical practice, significant **institutional variability** exists in treatment protocols which were not adjusted for in the model. Further validation in independent cohorts is necessary to confirm these findings, and strategies such as additional sub-group testing or external validation using larger independent cohorts should be employed.
- In our proposed model **severe disability at onset** (OR = 4.27, p = 0.044) and being **≥60 years old** at onset (OR = 4.25, p = 0.031) were the only **statistically significant markers** for predicting **poor outcomes (mRS ≥ 3)**. **Mood disorder** (OR = 0.14, p = 0.018), **psychosis** (OR = 0.13, p = 0.028) and **corticosteroid use** (OR = 0.08, p = 0.029) showed **protective effects**. This protective effect from the presence of mood disorder and psychosis is unexpected, warranting a further investigation of potential confounding variables. **Gait instability/ataxia**, **brainstem dysfunction**, and the **interval between symptom development and hospital admission** suggested trends towards poorer outcome, though they were **not statistically significant**.

ACKNOWLEDGEMENTS

A special thank you to Smaila Mulic, Adam Handel, Sophie Binks, the Autoimmune Encephalitis team at Nuffield Department of Clinical Neurology as well as the John Radcliffe Hospital, and the groups across the world contributing encephalitis cohorts for the final study.

GETTING IN TOUCH

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ENCEPHALITIS CONFERENCE 2024

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Results of Testing the Populations of Russia, Kazakhstan, and Armenia for Autoimmune Encephalitis Markers from 2019 to 2023

Dr Lev Brylev is a neurologist trained in Moscow, Russia. He has been the head of the Neurology Department in Buyanov city hospital for almost 10 years (from 2012 till 2022). During his work in this position his department became a reference centre for patients with multiple sclerosis, neuromyelitis optica and autoimmune encephalitis. Now he has moved to Abu Dhabi and works as consultant neurologist in ADSCC.

Results of Testing the Populations of Russia, Kazakhstan, and Armenia for Autoimmune Encephalitis Markers from 2019 to 2023

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Introduction

Autoimmune encephalitis (AE) is a rare disease, the diagnosis of which largely depends on the detection of specific antibodies in the blood and cerebrospinal fluid (1). The hypothesis of our study was that increasing the availability of testing would lead to higher detection rates of AE.

Methods

We analyzed an anonymized database of antibody test results for surface antigens from the largest reference laboratory "Invitro", which collects samples from the territories of Russia, Kazakhstan and Armenia for the period from 2019 to 2023. The total number of patients tested over these years is 4,421, of which 1,727 are men (41.0%) and 2,493 are women (59.1%). The average age was 36.5 years (SD 22 years), with 27.7% under 18 years old.

Results

The average age of patients with positive tests was 26.43 years (SD 15.95); 54 were men (26.9%) and 147 were women (73.1%), with 45.3% under 18. The positive group had a lower age and a higher proportion of women compared to whole tested group. The number of tests and positive results is shown in Table 1, with the highest positive rates for NMDAR and LGI1 antibody tests.

	Tested	Positive
AMPA1R_CSF	978	1
AMPA1R_S	1604	0
AMPA2R_CSF	977	0
AMPA2R_S	1604	3
CASPR_CSF	1063	0
CASPR_S	1063	4
GABAR_CSF	1879	2
GABAR_S	977	2
NMDAR_CSF	1571	95
NMDAR_S	3616	125
LGI1-CSF	1571	10
LGI1-S	1130	29

Table 1. Number of positive tests (S-serum, CSF –cerebrospinal fluid)

In the group of patients positive for LGI1 in blood or cerebrospinal fluid, the average age was 50.8 years (SD 19.4), with 55% being women. Patients positive for NMDA-R in cerebrospinal fluid had an average age of 18.4 years (SD 12.1), with 81.5% being women.

When evaluating the annual proportion of positive NMDA-R antibody tests, an increase in the number of tests and a decrease in the proportion of positive results in both blood and cerebrospinal fluid is notable (Table 2).

	2019	2020	2021	2022	2023
Total n NMDAR_CSF	112	243	316	423	477
Positive NMDAR_CSF	13	22	21	17	22
Positive (%)	10,4	8,3	6,2	3,8	4,4

Table 2. Proportion of positive tests from 2019 to 2023

With time, the proportion of patients undergoing paired blood and cerebrospinal fluid tests has increased, but the proportion of patients with double positive results in both blood and cerebrospinal fluid has also decreased. This indicates increased awareness among specialists and improved test availability.

An analysis was conducted on a group of patients who had both blood and cerebrospinal fluid tests for NMDA antibodies. The group included 835 people. Of the 45 patients with a positive blood test, 37 also had a positive cerebrospinal fluid test. Additionally, 24 patients had a positive cerebrospinal fluid test despite a negative blood test (Figure 1). This highlights the importance of performing cerebrospinal fluid analysis regardless of the blood test result.

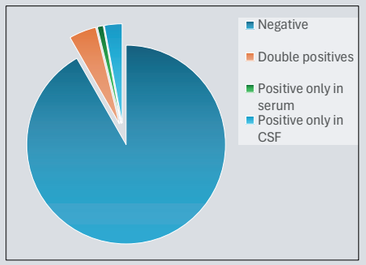

Figure 1. NMDA-R Ab in serum and CSF (n=835).

Discussion

The highest number of positive results was obtained from tests for NMDA-R and LGI1 receptor antibodies, which supports existing data in the literature (2). When resources are limited and other clinical data are lacking, these tests should be prioritized for suspected autoimmune encephalitis. It should be noted that the population of patients with a positive LGI1 test is significantly older than those with a positive NMDA-R antibody test. As previously reported (3) for autoimmune encephalitis with NMDA receptor antibodies, cerebrospinal fluid analysis significantly increases the likelihood of a positive test result, regardless of the blood test result.

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INVITRO

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Dr Marie Benaiteau

French Reference Centre on Paraneoplastic Neurological Syndromes and Autoimmune Encephalitis, Hospices Civils de Lyon, Hôpital Neurologique, Bron, France

Clinical spectrum and outcomes of cerebellar ataxia associated with glutamic acid decarboxylase antibodies

Dr Marie Benaiteau is a neurologist working at Professor Honnorat's French National Reference Centre for Autoimmune Encephalitis and Paraneoplastic Neurological Syndromes, in Lyon.

Clinical spectrum and outcomes of cerebellar ataxia associated with glutamic acid decarboxylase antibodies

Hanna Hutten[1,2], Marie Benaiteau[1,2] (MD), Macarena Villagrán-García[1,2] (MD), Géraldine Picard[1,2] (MSc), Chloé Buttard[1,2], David Goncalves[1,3] (PharmD), Véronique Rogemond[1,2] (PhD), Amna Klich[4] (PhD), Bastien Joubert[1,2] (MD PhD), Jérôme Honnorat[1,2] (MD PhD)

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Background: The clinical characteristics and long-term outcomes of patients with cerebellar ataxia and antibodies against the glutamic acid decarboxylase isoform 65 (GAD-Abs) are not well known.

Methods: This retrospective cohort study examined the clinical features and progression of patients with cerebellar ataxia and GAD65-Abs (GAD-CA) identified in the French Reference Centre on Paraneoplastic Neurological Syndromes and Autoimmune Encephalitis between January 1998 and December 2023.

Results:

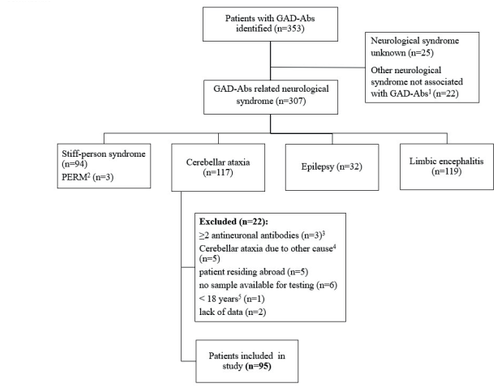

Figure 1 Study flowchart.
(1) Neuromyelitis optica (*n* = 1), neuropathy (n=6), upper motor neuron syndrome (*n* = 1), unspecified cognitive impairment (*n* = 1), post-stroke cognitive impairment (*n* = 2), HaNDL syndrome (*n* = 1), diplopia without other cerebellar symptoms (*n* = 1), progressive supranuclear palsey (*n* = 2), unspecified ambulatory problems (*n* = 2), Lewy-body dementia (*n* = 1), myelopathy (n=2), stroke (*n* = 2), Alzheimer's disease (*n* = 1). **(2)** One PERM patient had anti-GlyR antibodies, for the other two patients anti-GlyR antibodies were not tested. **(3)** anti-GABABr (*n* = 1), anti-CV2 (*n* = 1), anti-CASPR2 + anti-LGI1 (*n* = 1). **(4)** Post-neurosurgical (*n* = 1), SETX gene mutation (n=2), SCA type 2 (*n* = 1), CANVAS syndrome (*n* = 1). **(5)** A boy of 9 months old at onset of symptoms with a concurrent limbic encephalitis and IPEX syndrome.

Figure 3

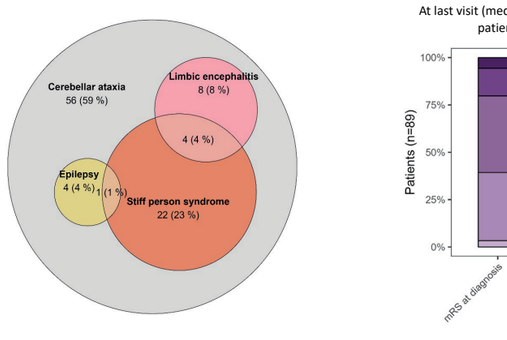

Figure 3 Venn-diagram of the overlap between GAD-Antibodies-spectrum disorders.

Figure 2

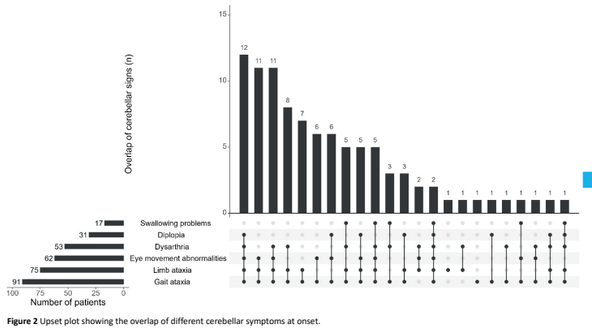

Figure 2 Upset plot showing the overlap of different cerebellar symptoms at onset.

Figure 4

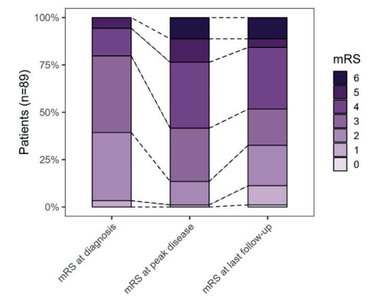

Figure 4 Evolution of modified Rankin Scale (mRS) at diagnosis, at the peak of the disease, and last follow-up (n=89).

Table 1 Patient characteristics — ***n* = 95**

Female	73 (76.8%)
Years of follow-up *median (min-max)*	3.0 (0-23)
Age of onset, years *mean*	58.0 (±12)
Other GAD-related syndromes	39 (41.1%)
Cancer	14 (14.7%)
Breast	6 (6.3%)
Type of onset	
Acute (< or = 7 days)	23/80 (28.8%)
Subacute (> 7 days – 3 months)	19/80 (23.8%)
Chronic (> 3 months)	38/80 (47.5%)
Unknown	15 (15.8%)
Paroxysmal signs pre-onset	18/66 (27.3%)
Diagnostic delay in months *median (min-max)*	12 (0-215)
Unknown	10
Therapeutic delay in months *median (min-max)*	12 (0-216)
Unknown	26
Gait ataxia at onset	91 (95.8%)
Limb ataxia at onset	75 (78.9%)
Ocular movement abnormalities at onset	62 (65.3%)
Dysarthria at onset	53 (55.8%)
Diplopia at onset	31 (32.6%)
Swallowing problems at onset	17 (17.9%)
Cognitive decline at any point	38 (40%)
Behavioural problems at any point	10 (10.5%)
Psychiatric problems at any point	25 (29.5%)
Other autoimmune disease	60 (63.2%)
Diabetes mellitus type 1/LADA	30 (31.6%)
Autoimmune dysthyroidism	35 (36.8%)
Pernicious anemia	8 (8.4%)
Other[1]	18 (18.9%)
Family history of autoimmune disease	24 (25.3%)
Inflammatory of the first CSF analyzed	59/89 (66.3%)
OCB	48/89 (53.9%)
Pleocytosis	21/89 (23.6%)
Cerebellar atrophy on MRI	28/88 (31.8%)
Inflammatory hyperintensities on MRI	13/88 (14.8%)
First-line immunologic therapy	81/91 (89%)
Intravenous immunoglobulins	78 (85.7%)
Corticosteroids	37 (40.7%)
Second-line immunologic therapy	57/91 (62.6%)
Rituximab	46 (50.5%)
Cyclophosphamide	38 (41.7%)
Azathioprine	13 (14.3%)
Mycophenolic acid	3 (3.3%)
Last follow-up, months *median (min-max)*	51 (1-286)
Progression between diagnosis and last follow-up	
Increased mRS (worsening)	37/89 (41.6%)
• Death	11 (11.6%)
Stable mRS (stability)	29/89 (32.6%)
Decreased mRS (improvement)	23/89 (25.8%)
Unknown	6

[1]Other autoimmune diseases: systemic lupus erythematosus (*n* = 1), cutaneous lupus erythematosus (*n* = 1), vitiligo (n=2), autoimmune myasthenia (*n* = 1), autoimmune neutropenia (*n* = 1), unspecified polyarthritis (*n* = 1), neurosarcoidosis (*n* = 1), lichen sclerosus (*n* = 1), eosinophilic granulomatosis with polyangiitis (*n* = 1), rheumatoid arthritis (*n* = 2), autoimmune hepatitis (*n* = 1), ANCA vasculitis (*n* = 1), latent adrenal insufficiency (*n* = 1), Sjögren's syndrome (*n* = 1), Celiac's disease (*n* = 1), psoriatic arthritis (*n* = 1).

- **No clinical, CSF or MRI features or immunoactive treatment were significantly associated with outcome.**
- However, patients with a mRS ≥3 at baseline more often present with an acute or subacute onset (63% versus 37,5%, p=0.079, *Chi2 test*).
- Patients with a mRS ≥3 6 months after the immunologic therapy have a longer diagnostic delay than the rest of the cohort (median 12 months versus 0.5 months, p=0.074 *Wilcoxon test*).

Conclusion : GAD-CA affects mostly middle-aged women with comorbid autoimmunity and remains a highly disabling disease. The variable mode of onset and inconstant CSF inflammation may explain the considerable diagnostic delay. Cognitive decline is observed in 40% of patients, warranting specific monitoring. Some patients with GAD-CA may responds to treatment. Large, prospective studies are needed to identify factors associated with disease activity.

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Serological and functional characterization of a cohort of patients with Seronegative Autoimmune encephalitis

Marta Mota, Msc is a second-year student at the master's program in Cellular and Molecular Biology at the University of Coimbra. Her master's project focuses on the pathophysiological mechanisms of Seronegative Autoimmune Encephalitis.

Serological and Functional Characterization of a Cohort of Patients with Seronegative Autoimmune Encephalitis

Marta Mota (1,2)*, João Moura (3)*, Mariana Ribeiro (1,2), Ana Luisa Carvalho (1,2), Ernestina Santos (3), UNITED* study & Ester Coutinho (1,2,4)

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(2) CiBB - Centre for Innovative Biomedicine and Biotechnology, University of Coimbra;
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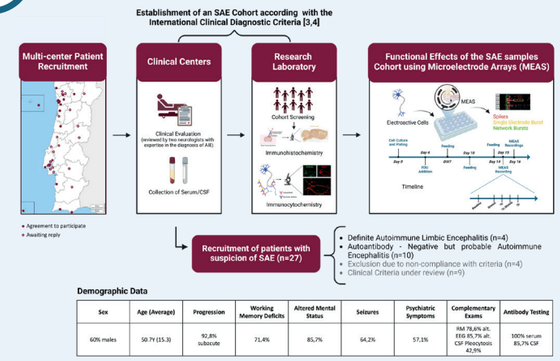
*Equal contribution

1 Introduction

- In recent years, research in the field of autoimmune encephalitis (AIE) has grown exponentially, and that led to the discovery of a multitude of different antigenic targets. *[1]*
- Nevertheless, there are some patients with the same clinical manifestations of AIE but lacking detectable autoantibodies in their serum or cerebrospinal fluid (CSF).
- This condition is designated as Seronegative Autoimmune Encephalitis (SAE), or antibody-negative AIE. [2]

Is SAE caused by autoantibodies to yet unknown antigenic targets or is it triggered by other pathophysiological mechanisms ?

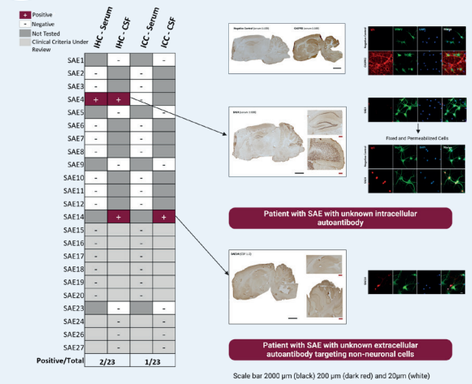
2 Study Design & Clinical Cohort



Demographic Data

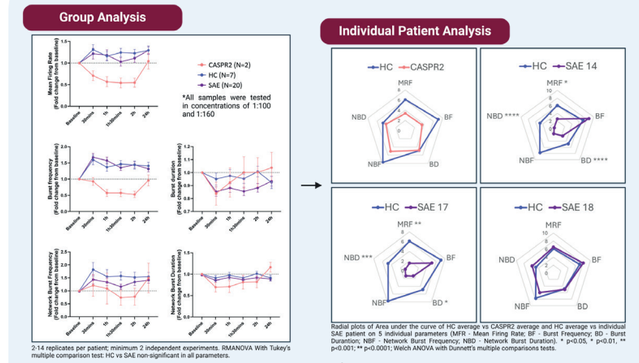
Sex	Age (Average)	Progression	Working Memory Deficits	Altered Mental Status	Seizures	Psychiatric Symptoms	Complementary Exams	Antibody Testing
60% males	50.7Y (15.3)	92,8% subacute	71,4%	85,7%	64,2%	57,1%	RM 78,6% alt. EEG 85,7% alt. CSF Pleocytosis 42,9%	100% serum 85,7% CSF

3 Cohort Screening on TBAs



2 patients with SAE clinical criteria had antibodies detectable by TBA

4 Cohort Screening on Functional MEA Assays



2-14 replicates per patient; minimum 2 independent experiments. RMANOVA With Tukey's multiple comparison test: HC vs SAE non-significant in all parameters.

- No changes in in vitro neuronal network activity upon exposure of murine cortical neurons to sera from the SAE cohort vs HC sera.

Radial plots of Area under the curve of HC average vs CASPR2 average and HC average vs individual SAE patient on 5 individual parameters (MFR - Mean Firing Rate; BF - Burst Frequency; BD - Burst Duration; NBF - Network Burst Frequency; NBD - Network Burst Duration). * p<0.05, * p<0.01, ** p<0.001; ** p<0.0001; Welch ANOVA with Dunnett's multiple comparisons tests.

- Significant changes in *in vitro* neuronal network activity upon exposure of primary cortical neurons to sera from 2/20 patients from the SAE cohort (SAE14 and SAE18) versus HC sera. The remainder of the cohort showed no differences from HC sera (illustrative example, SAE 18);
- One patient (SAE14) had antibodies towards unknown targets on TBAs.

5 Discussion/Conclusion

- Thus far, our data does not support the presence of a factor in serum, namely autoantibodies, as a pathogenic mechanism in the majority of patients with SAE.
- 2/23 patients (<10%) had antibodies detectable by traditional TBAs. 2/20 patient's sera (10%) had an electrophysiological functional effect on rodent cortical neuronal cultures. These are candidates for future antigen discovery.
- Future work will explore the effects of CSF from SAE patients and the relevance of other cell populations in this in vitro setting.

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Dr Massimiliano Ugo Verza

Department of Neurosciences, Psychology, Drugs Research and Child Health (NEUROFARBA), University of Florence, Florence, Italy

Isolated neuropathic pain in CASPR2 antibodies spectrum disease: case report and systematic review

Dr Massimiliano is currently working as Neurology resident at AOUC Careggi, Florence, where he is interested in antibody-mediated neurological conditions, with particular focus on Myasthenia Gravis and Autoimmune Encephalitis.

Isolated neuropathic pain in CASPR2 antibodies spectrum disease: case report and systematic review

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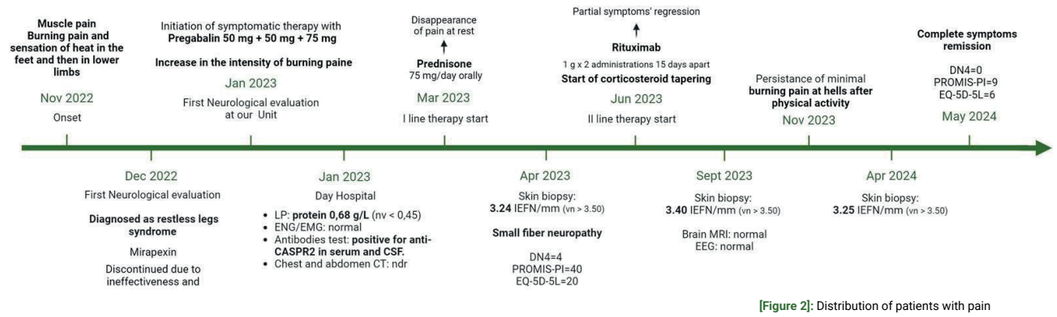
BACKGROUND

The spectrum of clinical features associated with contactin-associated protein-like 2 (CASPR2) antibodies includes acquired neuromyotonia, limbic encephalitis and Morvan syndrome. In this context, neuropathic pain has increasingly been reported as a feature of CASPR2 antibody spectrum disease, though data on its characteristics and prognosis remain limited. Here, we describe a patient with isolated neuropathic pain associated to CASPR2 antibodies and report the results from a literature review of similar cases to better define this unusual association.

CASE PRESENTATION

A 64-year-old male patient presented with rapidly progressive burning and tingling pain in his lower limbs. Neurological examination and electroneurography were normal, but a skin biopsy indicated small fiber neuropathy (3.24 IEFN/mm - normal range > 3.50 IEFN/mm). Diagnostic assessments for neuropathic pain revealed DN4 score of 4, PROMIS-PI score of 40, and EQ-5D-5L score of 20. Screening for neural antibodies led to the identification of CASPR2 antibodies in both CSF (1:2 dilution) and serum (1:100 dilution) samples by fixed and live cell-based assays. Brain MRI and electroencephalogram were unremarkable. Treatment with oral corticosteroids and gabapentin resulted in partial improvement, while the administration of rituximab as second-line treatment led to complete remission. At 19-month follow-up, the patient was asymptomatic, with improved clinical scores (DN4=0, PROMIS-PI=9, EQ-5D-5L=6), a skin biopsy was repeated resulting with 3.25 IEFN/mm
The patient's clinical history is summarized in the following timeline. [Figure 1]

[Figure 1]: Timeline of patient medical events.



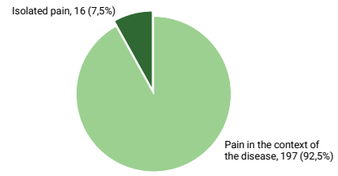
LITERATURE REVIEW

Literature research was performed on PubMed and EMBASE with the following search query: ["CASPR2" OR "VGKC"] AND ["neuropathy" OR "pain"]. The review was registered on PROSPERO and the PRISMA reporting guidelines were applied. Criteria of exclusions were: non original articles, editorials or conference abstract, articles text not available in English and animal studies. At the end of the review process 43 articles were kept for data extrapolation and analysis.

RESULTS

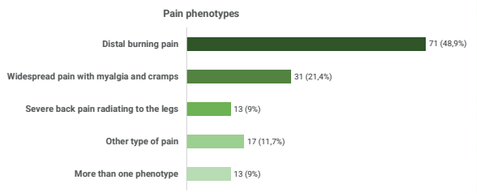
The literature review identified **213 patients with CASPR2 antibodies and neuropathic pain**. Neuropathic pain was often reported in association with other neurological symptoms of CASPR2 spectrum disease [Table 1], while **isolated pain** was described in only **16/213 (7,5%) cases** [Figure 2]. Pain was typically severe and had the main following characteristics: **distal burning pain** (48,9% and as in our patient), **widespread pain with myalgia and cramps** (21,4%), and **severe back pain radiating to the legs** (9%) [Figure 2B]. **Dysautonomic tests and skin biopsy** were reported in 19/213 and could detect small nociceptive fiber dysfunction in 16/19 [Table 2]. Data on **treatment response** were available in only 100/213 cases, showing complete or partial improvements of pain after immunotherapy in 82,1% of cases, though long-term immunotherapy or symptomatic medications are often required [Figure 4]. Objective **clinical scales** to measure outcomes were applied only in 14/43 studies, and they were mostly Modefied Rankin Scale and Numeric Rating Scale for pain.

[Figure 2]: Distribution of patients with pain



[Table 1]: Symptoms associated with pain

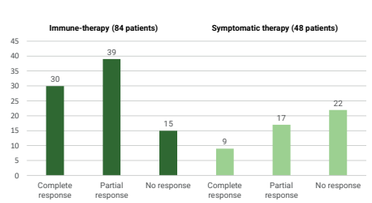
Symptoms associated with pain - No (%)	
Morvan syndrome	76 (62,8%)
Neuromyotonia	15 (12,5%)
Isaac's syndrome	8 (6,6%)
Central nervous system involvement	17 (14%)
Other	5 (4,1%)



[Figure 3]: Pain phenotype at presentation

Test performed at diagnosis and alterations	
ENG/EMG (71)	
Normal	12
Neuromyotonia or peripheral nerve hyperexcitability	38
Other aspecific alterations	21
Dysautonomic test or skin biopsy (19)	
Normal	3
Abnormal	16
CASPR2 antibodies essay of CSF (30)	
Negative	14
Positive	16

[Table 2]: Test performed at diagnosis



[Figure 4]: Therapy response

DISCUSSION AND CONCLUSION

Neuropathic pain appears to be a specific and relevant symptom in CASPR2 disease and, although rare, can be the solely clinical feature of the disease. Testing for CASPR2 antibodies should be included in the screening of patients with small fiber neuropathy as in these cases prompt immunotherapy is highly effective.

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ENCEPHALITIS CONFERENCE 2024

Miss Milene Hamdani

University of Liverpool, Medicine Department

A retrospective observational study: characterising adult patients with autoimmune encephalitis at a tertiary neurological centre

Building on her Biomedical Science degree from Kingston University in London, Miss Milene Hamdani cultivated a profound passion for scientific innovation and research. This drive led her to gain valuable experience in the biopharmaceutical industry. Subsequently, she was fortunate to secure a place at the University of Liverpool Medical School. Currently in her third year, she has developed a keen interest in neurological pathology and research. The diverse aetiology and complexity of encephalitis particularly captured my attention, prompting her to undertake a research project at the Walton Centre in Liverpool. This project focuses on characterizing adult patients with autoimmune encephalitis at this Tertiary Neurological Centre in the Northwest of England. She is optimistic that this research will provide deeper insights into the diagnostic and therapeutic implications of autoimmune encephalitis.

A Retrospective Observational Study: Characterising Adult Patients with Autoimmune Encephalitis at a Tertiary Neurology Centre

Milene Hamdani, Dr. Yun Huang, Professor Michael D Benedict

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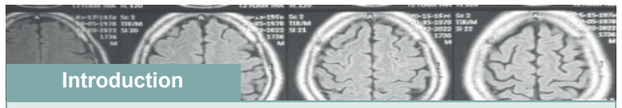

Introduction

Encephalitis is a severe inflammatory brain disorder caused by infectious and autoimmune aetiologies. While 40% of cases are of viral and idiopathic causes, 20% are attributed to immune origins where paraneoplastic sources are most common [1].

Autoimmune encephalitis encompasses a group of illnesses with varying pathophysiology marked by symptoms of limbic and extra-limbic debilitation [1]. It presents sub acutely with memory, cognition and behavioural deficits, psychiatric symptoms without fever or CSF pleocytosis, ensued by decreased consciousness and possibly coma [1, 2, 3].

Diagnosis has proven as challenging due to overlapping features of clinical presentation, laboratory and imaging findings between viral and autoimmune encephalitis [4]. While MRI and EEG prove useful to exclude other differentials and to manage seizures, no imaging methods are specific than the presence of neuronal antibodies for seropositive encephalitis [1]. Seronegative encephalitis heavily relies on the Graus Criteria for a diagnosis.

This research aims to identify the prevalence of patients admitted with seropositive and seronegative autoimmune encephalitis to better guide diagnosis and management.

Methods

A retrospective analysis was conducted on 92 patients using the EP2 database at the Walton Centre. These patients were residing in the North West of England and were either assessed or admitted at a secondary centre and later referred to the Walton Centre. Patients were categorised into three groups: seropositive, seronegative and negative controls who had another diagnosis.

Data Collected and Analysed:

- Demographic information
- Clinical presentation and history of tumours
- Laboratory findings (CSF analysis, autoantibody testing, sodium levels)
- Neuroimaging (MRI) and Electroencephalogram (EEG)
- Treatment options (Steroids, IVIg, plasma exchange or immunosuppressants)
- Glasgow Outcome Scale (GOS)

All data was non-parametric and statistical analysis using Kruskal-Wallis H test and Chi-squared test was performed.

Results

Differences in age showed no statistical significance (p = 0.376).

Seizures, altered mental status and confusion were prominent symptoms in both seronegative and seropositive cohorts. The negative cohort mostly had seizures, short term memory loss and movement disorders. Statistical significance across all three groups was found in patients presenting with fever, psychiatric disturbances, confusion, ataxia and signs of a movement disorder.

In the seropositive group, two patients had small cell lung cancer (SCLC) and one with thyroid cancer. There were no tumours diagnosed in the seronegative group. The negative cohort had one patient with a pseudo-inflammatory tumour, and another had a brain tumour.

Raised protein concentrations were mostly elevated in the negative group (41.3%, 18% and 15% in the control, seropositive and seronegative groups, respectively).
Oligoclonal bands were present in positive anti-NMDAR, LGI1 and GABA(b). No Type 2 bands were found in patients with IGI1 and seronegative encephalitis. There was no statistical significance of their presence across all three groups.
MRI showed hippocampal hyperintensities with volume loss and swelling limited to IGI1.
Hyponatraemia was largely found in IGI1 antibodies (57%) and in only one patient with seronegative encephalitis.

Autoantibodies were predominantly found in seropositive encephalitis. Most patients had LGI1 followed by NMDAR and GABA(b) antibodies. ANA was the sole antibody found in seronegative encephalitis, but with equal amounts across seropositive and seronegative groups. Most cases were in the negative group. LGI1 mostly presented with confusion, altered mental status, short term memory loss and seizures in comparison to other antibodies. Confusion, followed by seizures and altered mental status were dominant features of anti-NMDAR and focal neurology was not seen in any other antibody findings than NMDAR.

Steroids were more commonly used for seronegative encephalitis (85%) than seropositive encephalitis (66%) and IVIg. GOS showed no statistical difference (p = 0.364).

Table 1. Analysis of demographics and presenting symptoms.

	Seropositive (N = 44)	Seronegative (N = 20)	Control (N = 29)	P value
Demographics				
Age, years (IQR)	66.50 (54.50, 74.00)	68.50 (54.00, 74.75)	60 (47.00, 71.50)	0.376
Male, n (%)	27 (61.4)	9 (45.0)	14 (48.3)	0.369
Clinical Presentation on Admission, N (%)				
Fever	1 (2.3)	3 (15.0)	0	0.026
Seizures	20 (45.5)	13 (65.0)	11 (37.9)	0.166
Short-term memory loss	16 (36.4)	9 (45.0)	11 (41.4)	0.789
Psychiatric symptoms	9 (20.5)	10 (50.0)	2 (6.9)	0.002
Altered mental status	24 (54.5)	12 (60.0)	3 (10.3)	<0.001
Confusion	24 (54.5)	16 (80.0)	8 (27.6)	0.001
Language problems	10 (22.7)	7 (35.0)	8 (27.6)	0.587
Movement disorder	7 (15.9)	8 (40.0)	12 (41.4)	0.030
Ataxia	4 (9.1)	2 (10.0)	10 (34.5)	0.012
Bilateral external ophthalmoplegia	0	1 (5.0)	2 (6.9)	0.232
Past Medical History				
Depression	0	5	4	N/A
Psychosis	0	0	1	N/A

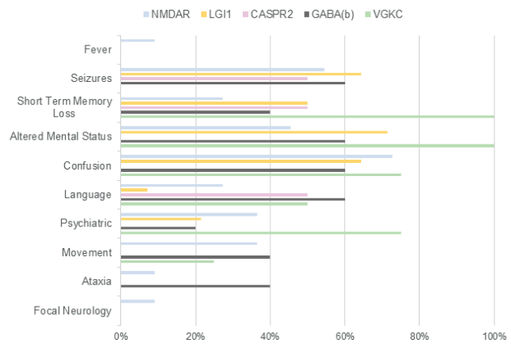

Figure 1. Proportion of antibody positive patients presenting with particular symptoms (%)

Discussion

Early clinical presentation can aid diagnosis. Anti-NMDAR encephalitis typically presents with confusion, altered mental status, and seizures. IGI1 presents similarly but no abnormalities in movement were found as per previous studies. Psychiatric changes with GABA(b) were less prominent, potentially due to low case numbers.

Oligoclonal bands and raised CSF protein were not helpful. Hyponatremia may differentiate seropositive and seronegative encephalitis. ANA may be a non-specific marker for autoimmune encephalitis, as it was present across all groups. MRI findings aligned with the Graus Criteria for seronegative encephalitis, though CSF pleocytosis data was insufficient.

No significant differences were found in GOS between seropositive and seronegative cohorts, suggesting similar clinical courses and treatment responses.

Conclusion

This report has shown clear differences in symptoms between seropositive and seronegative encephalitis compared to reported literature. While the gross findings of seronegative encephalitis fall in line with the 2016 Graus Criteria guidelines, they are specific, thus adjustments may be required for subpopulation groups living in the North West of England.

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Dr Milena Trentinaglia

Neurology Unit, Department of Neurosciences, Biomedicine, and Movement Sciences, University of Verona, Verona, Italy

Epidemiology and clinical characteristics of autoimmune encephalitis in the province of Verona, Italy

Dr Milena Trentinaglia is a current 3rd-year Resident in Neurology at University of Verona, Italy. During her last year of medicine and her resident training she had the possibility to cultivate her interest in the field of Neuroimmunology. In particular, she was able to follow outpatients visit of neuroimmunological patients and learn the basic interpretation of laboratory diagnostic techniques and she keeps trying to deepen her knowledge in this fascinating field.

EPIDEMIOLOGY AND CLINICAL CHARACTERISTICS OF AUTOIMMUNE ENCEPHALITIS IN THE PROVINCE OF VERONA, ITALY

Milena Trentinaglia*[1], Alessandro Dinoto*[1], Sara Carta[1], Sergio Ferrari[1], Riccardo Tiberi[2], Bruno Bonetti[3], Federica Vit[4], Francesca Rossi[5], Alberto Cossu[6], Simone Fusina[7], Ava Easton[8,9], Sara Mariotto[1] on the behalf of Encephalitis International

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AIM OF THE STUDY

To investigate the **epidemiology and clinical features of antibody-positive autoimmune encephalitis (AE) and its mimics in the province of Verona**, Italy.

MATERIALS AND METHODS

We retrospectively selected patients residing in the province of Verona at the time of testing, referred for **antibody testing for suspected AE** to our Neuropathology Laboratory **from January, 1st 2020 to December, 31st 2023.**

All patients fulfilling 2016 criteria for "possible AE", "definite AE", "definite limbic encephalitis", and "antibody negative but probable AE" were included. Clinical and paraclinical data were collected. Cases were reviewed and classified as AE or as mimics under the supervision of a neurologist with experience in autoimmune encephalitis. ISTAT population data were used to estimate the incidence of AE and its mimics (population: 926.497 inhabitants)

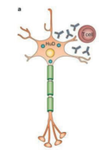
CLINICAL RESULTS

A **total of 330 patients** were identified during the study period and **92 of them fulfilled criteria for "possible AE"[3]** and were thus included for further analysis.

Among those patients, representing potential mimics of AE, final diagnoses were inflammatory (n=24, 26.1%), epileptic (n=23, 25%), infectious (n=18, 19.6%), neurodegenerative (n=9, 9.8%) and neoplastic (n=7, 7.6%) conditions. Less represented phenotypes were metabolic (n=4, 4.3%), vascular (n=4, 4.3%) and rheumatologic (n=1, 1.1%).

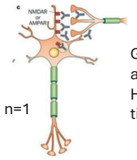
Additionally, "Definite AE" was diagnosed in 14 patients, all antibody-positive and with consistent clinical phenotype (Figure 1).

NEURAL SURFACE ANTIBODIES



NMDAR n=2
LGI1 n=3
CASPR2 n=1
CASPR2 and LGI1 n=1
GABAaR n=1
mGluR1-like IHC pattern n=1
MOG n=1

ANTIBODIES TO INTRACELLULAR ANTIGENS



GAD-65 n=1
amphiphisin n=1
Hu n=1
titin and CV2 n=1

Figure 1 – Proportion of antibody types in the studied cohort.

PROVINCE OF VERONA, ITALY
2020-2023
3.8 cases/million/ year

PROVINCES OF TRENTO AND TREVISO, ITALY[1]
2013-2018
1.54 cases/milion/ year

FRANCE[2]
2016-2018
3.6 cases/milion/ year

Figure 2 – Comparison of estimated incidence of antibody-positive AE in previous studies.

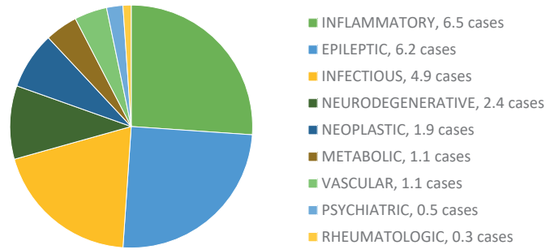


Figure 3 – Final diagnosis in patients fulfilling criteria for possible AE and estimated incidence (nr of cases/million/year)

EPIDEMIOLOGICAL RESULTS

We observed that the **estimated incidence of antibody-positive AE in the province of Verona was 3.8 cases/1.000.000 inhabitants/year**, which was comparable with previous studies reporting the epidemiology of AE (Figure 2).

Estimated incidence for the different AE mimics is reported Figure 3. Other inflammatory CNS disorders and epilepsy were the most frequently encountered mimics. Overall, **the incidence of all AE mimics was 24.9 cases/1.000.000 inhabitants/year.**

CONCLUSIONS

Our study suggests that, on an epidemiological basis, **potential AE mimics are 6 times more frequent than true cases of AE**, prompting caution in the diagnosis of AE and strict exclusion of other causes.

The alternative diagnoses of potential mimics were different from those reported in previous literature[4,5], reflecting significant differences in study population and design (retrospective studies from reference centers versus population-based studies).

The similar incidence of AE in our study compared to the literature suggests that our findings may be generalizable, however further studies are needed to confirm our findings.

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Disclosures: the present study was funded by Encephalitis International (Seed funding 2023 Italy).

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ENCEPHALITIS CONFERENCE 2024

Dr Niels Vander Elst

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Bacteriophage-encoded endolysins as innovative antimicrobials for the future treatment of pneumococcal meningoencephalitis

Dr Niels Vander Elst is a postdoctoral researcher affiliated to the Karolinska Institute, Sweden. He specializes in engineering endolysin variants with applications in (veterinary) medicine and beyond. Dr Vander Elst earned a dual PhD in Biotechnology and Veterinary Medicine that was jointly awarded by Ghent University and KU Leuven, Belgium. During his doctoral research, he spent a significant amount of time working on endolysins at the Institute for Bioscience and Biotechnology Research (IBBR) associated to the University of Maryland, Rockville, MD, USA. Dr Vander Elst has received recognition through scholarships and fellowships, also holding inventorship to one provisional and one full patent application.

Bacteriophage-encoded endolysins as innovative antimicrobials for the future treatment of pneumococcal meningoencephalitis

Niels Vander Elst[1], Lisa Knörr[1], Miguel T. Vian[1], Anders P. Håkansson[2] and Federico Iovino[1]

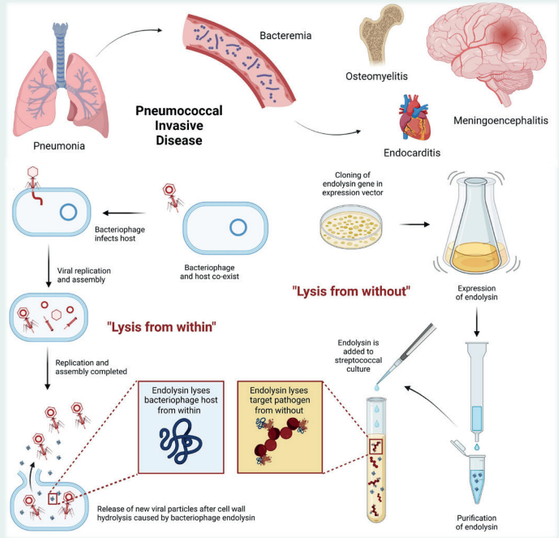
[1]Department of Neuroscience, Karolinska Institutet, Stockholm, Sweden.
[2]Department of Translational Medicine, Faculty of Medicine, Lunds Universitet, Malmö, Sweden.

Conclusions

- Endolysins CPL-1 and CPL-7s display high killing efficacy against a broad panel of *Streptococcus pneumoniae* isolates, including penicillin-resistant strains, all originating from patients with pneumococcal invasive disease.
- The endolysin killing activity is retained in human cerebrospinal fluid and blood.
- Endolysins kill penicillin-resistant pneumococci in the presence of neuronal-like cells and mitigate neurotoxicity.

Introduction

What is pneumococcal meningoencephalitis and how can endolysins help?



Pneumococci can enter the brain, meninges and other areas of the human body following bacteremia that originates from the respiratory tract. / The action of endolysins in a 'lysis-from-within' *versus* 'lysis-from-without' scenario is demonstrated. Bacteriophages employ endolysins at the end of their lytic replication cycle to release newly assembled viral particles from the infected host, causing 'lysis-from-within'. In the 'lysis-from-without' scenario, the endolysin gene is cloned in a vector, after which the endolysin is expressed and purified. In the case of Gram-positive pathogens, the purified endolysin retains its functionality when applied externally to the targeted pathogen (created with https://biorender.com/).

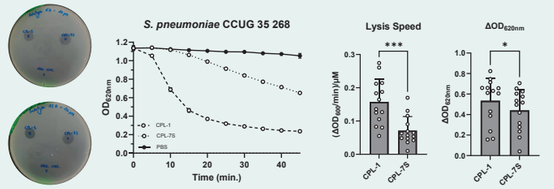
Bacterial meningoencephalitis, a severe infection of the brain and meninges, is primarily caused by *Streptococcus pneumoniae*. It is a major global health concern, especially in children, that often results in high mortality and lifelong neurological damage. With antimicrobial resistance on the rise, attention has turned to innovative approaches like endolysins derived from bacteriophages. These enzymes degrade bacterial cell walls, offering a promising alternative for combating antimicrobial resistant infections.

Methods

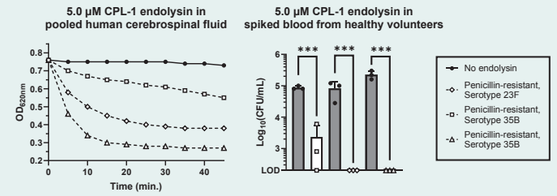
Two endolysins, CPL-1 and CPL-7s, chosen for their dimeric and monomeric characteristics respectively, were tested using enzyme-specific assays against a diverse panel of clinical strains, including antibiotic-resistant isolates. SH-SY5Y neuroblastoma cells were differentiated into mature neurons, infected and treated with endolysin, while kinetically monitoring bacterial growth (by plating) and cell death (by measuring lactate dehydrogenase levels in cell culture supernatant).

Results

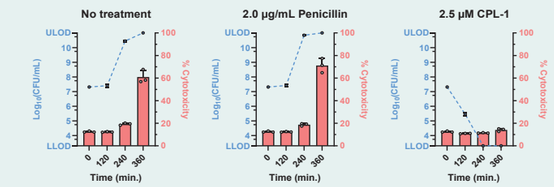
In enzyme-specific assays, the CPL-1 and CPL-7s endolysins displayed lytic activity against all tested pneumococcal isolates, with CPL-1 outperforming CPL-7s.



CPL-1 endolysin retains its lytic activity against penicillin-resistant isolates in pooled human cerebrospinal fluid and spiked blood from healthy volunteers.



CPL-1 endolysin kills penicillin-resistant pneumococci in the presence of neuronal-like cells and mitigates neurotoxicity.



Dr. Vander Elst is a postdoctoral researcher affiliated to the Karolinska Institute, Sweden. He specializes in engineering endolysin variants with applications in (veterinary) medicine and beyond. Dr. Vander Elst earned a dual PhD in Biotechnology and Veterinary Medicine that was jointly awarded by Ghent University and KU Leuven, Belgium. During his doctoral research, he spent a significant amount of time working on endolysins at the Institute for Bioscience and Biotechnology Research (IBBR) associated to the University of Maryland, Rockville, MD, USA.

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Dr Pietro Businaro

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Serological quantitative, qualitative and functional profiling and neurofilaments as biomarkers in autoimmune encephalitis with LGI1 and CASPR2 antibodies

Dr Pietro Businaro is a neurologist and PhD student with a focus on antibody effector function in autoimmune neurology. Currently, he is a Neuroimmunology consultant at the IRCCS Mondino Foundation. His clinical expertise is rooted in significant experience at both the IRCCS Neurological Institute Carlo Besta and the IRCCS Neurological Institute Casimiro Mondino. Complementing his clinical work, he has nearly two years of preclinical experience in Neuro-oncology in Prof. Locatelli's lab at Bambino Gesù Children's Hospital. Over the past year, he has focused on the project "Prognostic relevance of quantitative and longitudinal CASPR2 and LGI1 antibody testing and neurofilament light chain levels in patients with autoimmune encephalitis" at the Neuroimmunology lab of the IRCCS Mondino Foundation. From July to mid-October, he expanded his research horizons as a fellow in the neuroimmunology lab of Prof. Dalmau and Prof. Graus in Barcelona under the supervision of Dr Marianna Spatola, focusing on identifying optimal diagnostic tests for paraneoplastic neurological syndromes associated with intracellular antibodies.

FONDAZIONE MONDINO
Istituto Neurologico Nazionale a Carattere Scientifico | IRCCS

UNIVERSITÀ DEGLI STUDI DI PAVIA
DIPARTIMENTO DI SCIENZE DEL SISTEMA NERVOSO E DEL COMPORTAMENTO

International Workshop on Antibody-Mediated Neurological Diseases
19 October 2024
Rome, Italy

Serological quantitative, qualitative and functional profiling and neurofilaments as biomarkers in autoimmune encephalitis with LGI1 and CASPR2 antibodies

Businaro P.[1,2], Masciocchi S. [1,2], Barnabei R. [1], Scaranzin S. [2]; Morandi C. [2], Bernini S. [3], Barone P. [4], Torriello A. [4], Arbasino C. [5], Benedetti L. [6], Cerne D. [7], Volonghi I. [8], Casagrande S. [9], Nassani S. [10], Currò Dossi M. [11], Micheli S. [12], Fattapposta F. [13], Leone C. [14], Risi M. [15], Paoletti M. [16], Marchioni E. [17], Franciotta D. [2], Gastaldi M. [2]

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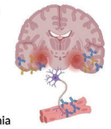
Introduction

Disease spectrum	LGI1-IgG	CASPR2-IgG
Autoimmune encephalitis		
Morvan Syndrome		
Neuromyotonia		



Pathogenic mechanism[1,2]	LGI1-IgG	CASPR2-IgG
Complement activation	not studied	yes
Antibody dependent cytotoxicity	not studied	yes
Antibody dependent cell phagocytosis	not studied	yes
Receptor internalization	not studied	yes
Protein-protein disruption	yes	yes

Literature Gap

Complement activation and innate immunity's pathogenic roles are relevant in CASPR2-associated disease (AD) but unexplored in LGI1-AD. Neurofilament light chain levels, IgG subclasses and titers are promising biomarkers in CASPR2/LGI1-AD.

Aim

To explore pathogenic mechanisms in CASPR2/LGI1-AD and study the relevance of longitudinal CASPR2/LGI1-IgG titers, subclasses and neurofilament light chain levels as biomarkers.

Materials and Methods

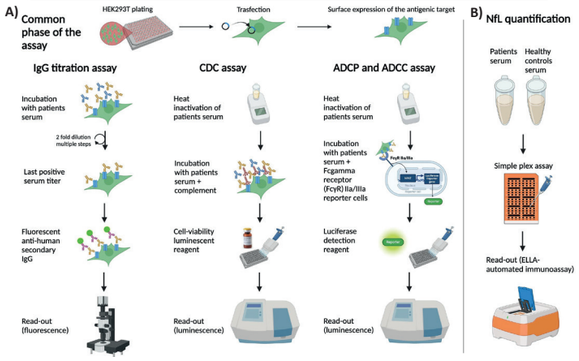


Figure 1. Description of A) IgG-titration, Complement-dependent cytotoxicity (CDC) and Antibody-dependent cytotoxocity (ADCC) and phagocytosis (ADCP) assays. B) Neurofilament light chain (NfL) quantification.

Results – I

Acute phase samples (n=22) had higher median CASPR2/LGI1-IgG titers (1:10240, range 1:20-1:102400) compared to remission samples (n=98) (1:800, range 0-1:51200, p<0.001) **(panel A)**. CASPR2/LGI1-IgG titers in patients with follow-up samples (n=19) decreased over time, being significantly higher at onset vs T4 (p<0.001), T10 (p=0.004), and last follow-up (p=0.009) **(panel B)**. Relapses always occurred with a positive sample (n=10) **(panel C)**. Seven patients seroreverted (2 with CASPR2-AD) after a median of 12 months (range 5-58), and none experienced post-seroreversion relapse (median follow-up 12 months, range 0-25) (not shown).

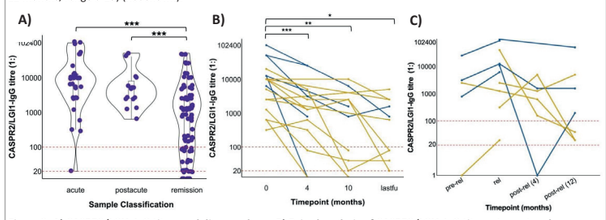


Figure 2. A) CASPR2/LGI1-IgG titers and disease phase. B) Paired analysis of CASPR2/LGI1-IgG titers at onset and longitudinal remission timepoints. C) CASPR2/LGI1 IgG titers and relapses. CASPR2 data are shown in blue, LGI1 data are shown in yellow, CASPR2/LGI1 pooled are shown in purple

NfL levels (median: 45.95, range 5.4-109 pg/ml) were higher (p<0.001) than in age/sex-matched healthy controls (median: 14.55, range 5.4-56.8 pg/ml) **(panel A)** and reduced at last follow-up compared to onset (median reduction: 20.25 pg/mL, range -9.3-88.7, p=0.03) but remained higher than controls (p=0.02) **(panel B)**. NfL levels at onset significantly correlated with a lower MoCA score at follow-up (rho=-0.672,p=0.02) confirmed by univariate linear regression (B=-3.881, p=0.0256) **(panel C)**.

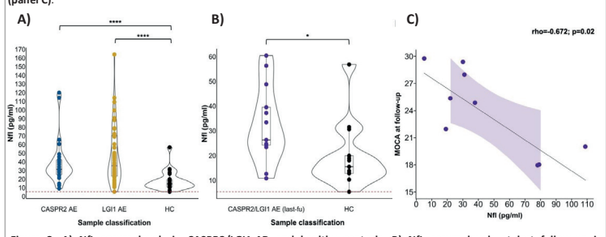


Figure 3. A) Nfl serum levels in CASPR2/LGI1-AD and healthy controls. B) NfL serum levels at last follow-up in CASPR2/LGI1-AD versus healthy controls. CASPR2/LGI1 Nfl levels at onset and MOCA at follow-up. CASPR2 data are shown in blue, LGI1 data are shown in yellow, CASPR2/LGI1 pooled are shown in purple.

Results - II

At onset all patients (n=19) had IgG4, which were the only subclass in only 2 (with CASPR2-IgG), Sixteen had additional IgG1 (n=3), IgG3 (n=5) or both (n=8). Seventeen/18 patients were still positive for the IgG4 subclass at last positive sample available, with 7/17 being exclusively IgG4 positive (3 CASPR2, 3 LGI1, one double positive) and 6 with other concomitant subclasses at onset became IgG4 positive only (1 CASPR2 and 5 LGI1) **(panel A)**. No CDC was observed in 23 patients (15 LGI1-IgG, 7 CASPR2-IgG, 1 CASPR2/LGI1-IgG), despite 20/23 being IgG1, IgG3, or IgG1/3 positive **(panel B)**.

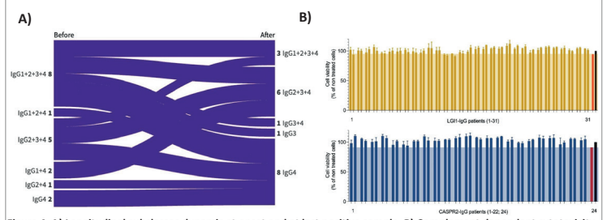


Figure 4. A) Longitudinal subclasses dynamic at onset and at last positive sample. B) Complement dependent cytotoxicity in LGI1-IgG patients (upper panel) and CASPR2-IgG patients (lower panel). CASPR2 data are shown in blue, LGI1 data are shown in yellow, CASPR2/LGI1 pooled are shown in purple

ADCC was found in 17/25 CASPR2-IgG samples and 9/33 LGI1-IgG samples. ADCP was present in 21/25 CASPR2-IgG samples and 20/33 LGI1-IgG samples.

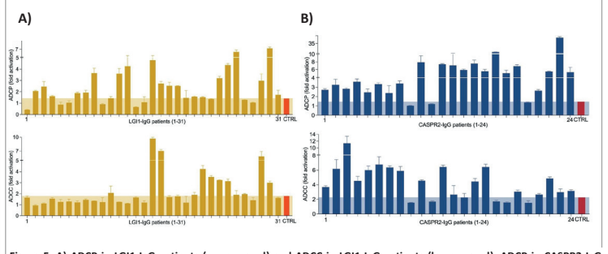


Figure 5. A) ADCP in LGI1-IgG patients (upper panel) and ADCC in LGI1-IgG patients (lower panel). ADCP in CASPR2-IgG patients (upper panel) and ADCC in CASPR2-IgG patients (lower panel).

Conclusion

CASPR2/LGI1-IgG titres correlate with disease phase, and seroreversion might help stratify patients at lower relapse risk. Conversely, high NfL levels might identify patients at higher risk of long-term cognitive disability. We identified antibody-mediated innate immunity activation as a prominent disease mechanism compared to complement activation in LGI1/CASPR2-AD, suggesting potentially novel treatment strategies.

References

1.Papi, C., Milano, C. & Spatola, M. Mechanisms of autoimmune encephalitis. Curr. Opin. Neurol. 37, 305–315 (2024). 2.Terroba-navajas, P., Spatola, M. & Chuquisana, O. Article Humoral signatures of Caspr2-antibody spectrum disorder track with clinical phenotypes and outcomes Humoral signatures of Caspr2-antibody spectrum disorder track with clinical phenotypes and outcomes. (2025) doi:10.1016/j.medj.2024.09.004.

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Softwares and tools

Figure and statistical analysis were performed with R studio, graphpad and Biorender.com

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Dr Pietro Businaro

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Commercial tissue-based assays are suboptimal to detect antibodies against intracellular neuronal antibodies in autoimmune neurological syndromes

Dr Pietro Businaro is a neurologist and PhD student with a focus on antibody effector function in autoimmune neurology. Currently, he is a Neuroimmunology consultant at the IRCCS Mondino Foundation. His clinical expertise is rooted in significant experience at both the IRCCS Neurological Institute Carlo Besta and the IRCCS Neurological Institute Casimiro Mondino. Complementing his clinical work, he has nearly two years of preclinical experience in Neuro-oncology in Prof. Locatelli's lab at Bambino Gesù Children's Hospital. Over the past year, he has focused on the project "Prognostic relevance of quantitative and longitudinal CASPR2 and LGI1 antibody testing and neurofilament light chain levels in patients with autoimmune encephalitis" at the Neuroimmunology lab of the IRCCS Mondino Foundation. From July to mid-October, he expanded his research horizons as a fellow in the neuroimmunology lab of Prof. Dalmau and Prof. Graus in Barcelona under the supervision of Dr Marianna Spatola, focusing on identifying optimal diagnostic tests for paraneoplastic neurological syndromes associated with intracellular antibodies.

IDIBAPS

UNIVERSITÀ DEGLI STUDI DI PAVIA
DIPARTIMENTO DI SCIENZE DEL SISTEMA NERVOSO E DEL COMPORTAMENTO

International Workshop on Antibody-Mediated Neurological Diseases
19 October 2024
Rome, Italy

Commercial tissue-based assays are suboptimal to detect antibodies against intracellular neuronal antibodies in autoimmune neurological syndromes

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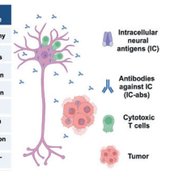
Introduction

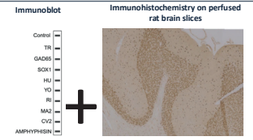
Autoimmune disorders with IC-Abs

IC-abs	Tumor risk	Clinical syndrome
HU	High	Neuronopathy
RI	High	Brainstem encephalitis
YO	High	Cerebellar degeneration
TR	High	Cerebellar degeneration
GAD65	Low	Epilepsy
AMPHI	High	Stiff person syndrome
SOX1	High	Lambert eaton MS
CV2	High	Encephalo - Myelitis
MA2	High	Limbic encephalitis
GFAP	Low	Encephalo - Myelitis



Clinical relevance: diagnosis, prognosis and tumor search

How to test IC-abs



Only concordant results between tissue-based assays and immunoblots are associated with paraneoplastic neurological syndrome

Literature Gap

Many diagnostic laboratories use commercial immunofluorescence tissue-based assays (IIF-TBAs) to detect antibodies against intracellular antigens (IC-abs) for which the diagnostic value is yet to be determined

Aim

To compare the performances of two commercial IIF-TBAs (INOVA and EUROIMMUN) in detecting IC-abs

Materials and Methods

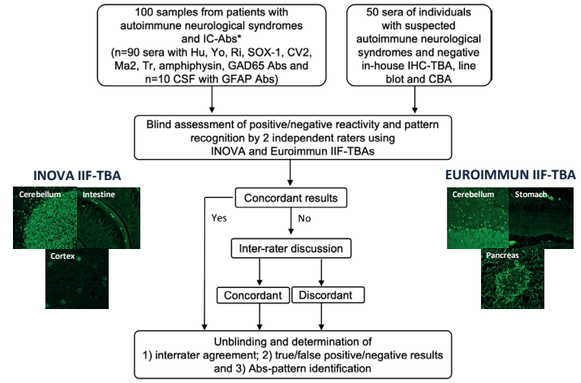
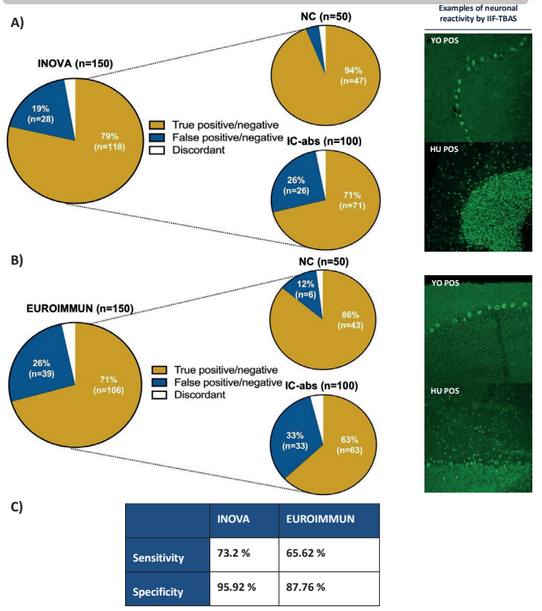


Figure 1. Algorithm of the study. *confirmed by in-house TBA and immunoblot and/or Cell-based assay

Results - I



	INOVA	EUROIMMUN
Sensitivity	73.2 %	65.62 %
Specificity	95.92 %	87.76 %

Figure 2. Proportion of correctly identified (yellow) and misclassified (blue) samples, overall (left pies) and when considering only positive (IC-abs) or negative controls (NC) (right pies) with INOVA (A) and EUROIMMUN (B) IIF-TBAs. Overall statistics performances (C).

Results - II

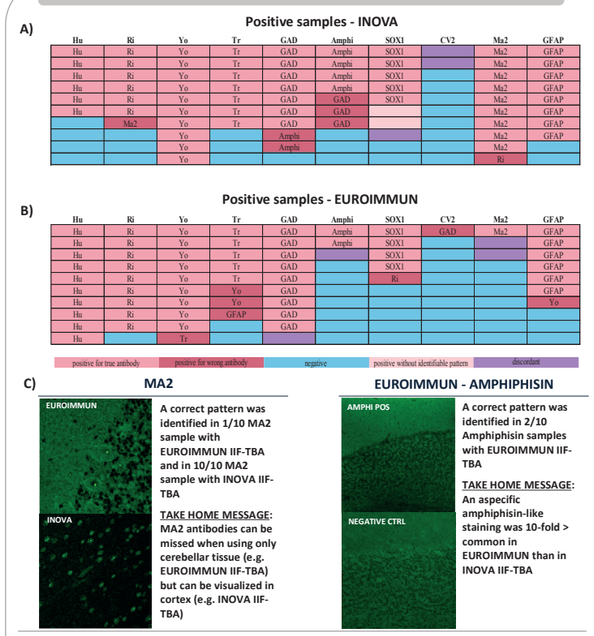


Figure 3. Heatmap illustrating the detailed results for each sample tested with INOVA (A) and EUROIMMUN (B). Each row correspond to one sample, each column to one IC-ab. **Pitfalls in pattern identification in INOVA and EUROIMMUN IIF-TBAs (C).**

Conclusion

Diagnostic performance of both commercial IIF-TBAs to identify IC-Abs is suboptimal. Commercial IIF-TBAs can miss rare (amphyphisin, CV2 and MA2) and less rare antibodies (HU) and should not be used as screening test for detection of IC-abs.

References

Ruiz-García R, Martínez-Hernández E, Saiz A, Dalmau J, Graus F. The Diagnostic Value of Onconeural Antibodies Depends on How They Are Tested. *Front Immunol*. 2020;11(July):1-6. doi:10.3389/fimmu.2020.01482

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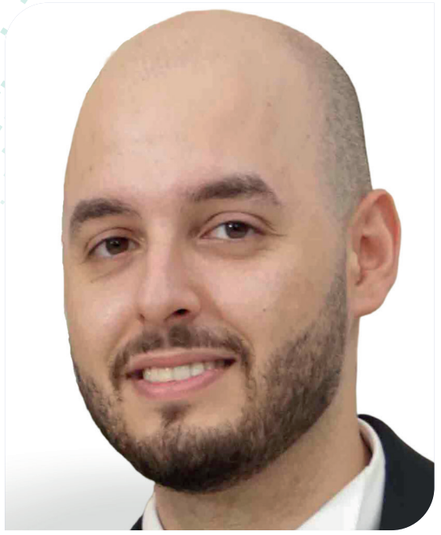


Dr Ralph Habis

Johns Hopkins Encephalitis Center, Department of Neurology, Johns Hopkins University School of Medicine, Maryland, USA

Comparative Analysis of Clinical Characteristics, Hospitalization Burden, and Costs of Autoimmune Encephalitis and HSV Meningo-Encephalitis

Dr Ralph Habis earned his MD from the American University of Beirut in June 2022, and since August 2022, he has been conducting a post-doctoral research fellowship at the Johns Hopkins Encephalitis Center and Adult Epilepsy Diet Center.

Comparison of Clinical Characteristics, Hospitalization Outcomes, and Costs of Autoimmune Encephalitis and HSV Meningoencephalitis

Ralph Habis, MD[1]; Arun Venkatesan, MD, PhD[1] ; John Probasco, MD[1]

[1]Johns Hopkins Encephalitis Center, Department of Neurology, Johns Hopkins University School of Medicine, Maryland, USA

Introduction

- Encephalitis imposes a substantial burden on healthcare systems with costs estimated at $2 billion in the United States alone in 2010 and a global incidence surpassing 1 million in 2019.
- Autoimmune encephalitis as a significant encephalitis type, though many who meet clinical consensus criteria are seronegative for an associated antibody in serum and CSF.
- Comparative studies on the hospitalization burden of various encephalitis types and the factors contributing to this burden are lacking.

Objectives

- Delineate the differences in hospitalization costs and outcomes between cohorts of seronegative, seropositive autoimmune encephalitis (AIE) and herpes simplex virus meningoencephalitis (HSE).
- Explore factors contributing to increased length of stay (LOS) and hospital charges within these distinct cohorts.

Materials and Methods

- **Study Population:** Identified by querying billing databases for discharge diagnoses of encephalitis at Johns Hopkins Hospital and Johns Hopkins Bayview Medical Center in Baltimore, MD (using ICD-9-CM codes) from March 1, 2014, to June 30, 2022.
- **Data Collection:** Retrospectively from the EMR.
- **Outcome Measures:** Change in the modified Rankin Scale (mRS) from admission to discharge, hospital charges, hospitalization LOS, the need for ICU admission, discharge disposition, and mortality.
- **Statistical Analysis:**
  - Bivariate Analysis (Chi-square, t-test) to:
    - Compare clinical characteristics and outcome measures between two cohorts, seropositive or seronegative AIE vs. HSE.
    - Subgroup analysis to compare seronegative vs. seropositive AIE.
    - Identify factors associated with a LOS>10 days in each type of encephalitis.
  - Multivariate linear regression: explore contributors to increased hospital charges.

Results

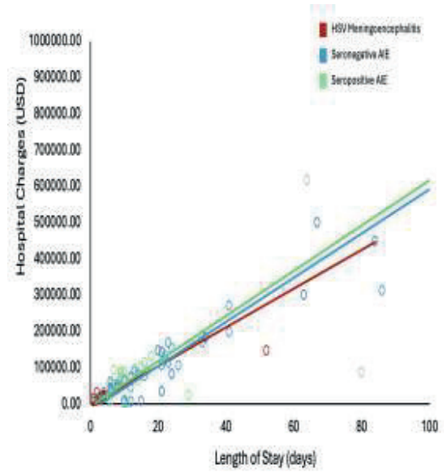
Comparing Hospitalization Burden of Autoimmune Encephalitis and Herpes Simplex Meningoencephalitis

	Autoimmune Encephalitis 74/111 (66.7%)	Herpes Simplex Meningoencephalitis 37/111 (33.3%)	P-Value
ICU Admission	25/73 (34.2)	10/37 (27)	0.44
Mortality	2/74 (2.7)	2/37 (5.4)	0.6
Median Length of Stay at The Hospital	11.5 days (IQR: 8-22.8)	5 days (IQR: 3-22.5)	**0.01**
Change in mRS-Admission to Discharge :			0.23
Improved (by >1)	30/61 (49.1)	18/37 (48.6)	
No change	17/61 (27.9)	15/37 (40.5)	
Worsen (by < 1)	14/30 (23)	4/37 (10.8)	
Median Hospital Charges (USD)	$71,735 (IQR: $42,589 - $116,753)	$20,619 (IQR: $13,951-$81,455)	**0.007**
Disposition:			**0.001**
Home	42/72 (58.3)	19/37 (51.4)	
Home with care	5/72 (6.9)	7/37 (18.9)	
Skilled nursing	2/72 (2.8)	7/37 (18.9)	**0.004**
Rehab	20/72 (27.8)	1/37 (2.7)	**0.002**
Hospice	0/72	1/37 (2.7)	
Other	3/72 (4.2)	2/37 (5.4)	

Comparing Inpatient Treatment and Burden of Seronegative and Seropositive AIE

	Seronegative AIE 41/74 (55.4%)	Seropositive AIE 33/74 (44.6%)	P-Value
Steroids	37/41 (90.2%)	28/33 (85)	0.48
IVIg	14/41 (34)	15/33 (45.5)	0.32
PLEX	11/41 (27)	18/33 (54.5)	**0.01**
Combination of First Line	17/41(41.5)	22/33 (66.7)	**0.03**
Need for second line	3/36 (8.3)	10/33 (30.3)	**0.02**
ICU Admission	15/41 (36.6)	10/33 (31.2)	0.63
Mortality	2/41 (5%)	0	0.2
Median Length of Stay at The Hospital	10 days (IQR: 8-18)	13 days (IQR: 6-26)	0.49
Change in mRS-Admission to Discharge:			0.67
Improved (by >1)	15/30 (50)	15/31 (48.4)	
No change	7/30 (23.3)	10/31 (32.3)	
Worsen (by < 1)	8/30 (26.7)	6/31 (19.4)	
Median Hospital Charges (USD)	$71,798 (IQR: $53,472- $96,291)	$71,735 (IQR: $28,616-$167,674)	0.95
Disposition:			0.91
Home	24/41 (58.5)	18/33 (54.5)	
Home with care	2/41 (4.9)	3/33 (9.1)	
Skilled nursing	1/41 (2.4)	1/33 (3)	
Rehab	12/41 (29.4)	8/33 (24.3)	
Hospice	0	0	
Other	2/41 (4.8)	3/33 (9.1)	

Scatterplot of Total Hospital Charges vs. Hospital Length of Stay



Factors Associated with a Prolonged Length of Stay

Variable	Hospital Length of Stay>10 days	Hospital Length of Stay ≤ 10 days	p-value
Herpes Simplex Encephalitis N=37			
Median Age	61 (IQR: 48-64)	37 (IQR: 29-59)	**<0.001**
CCI≥2	12/14 (85.7)	4/23 (17.4)	**<0.001**
Immunosuppressed	11/14 (78.6)	3/23	**<0.001**
Altered Mental Status	13/14 (93)	3/23 (13)	**<0.001**
Focal Neurological Deficits	9/14 (64.3)	4/23 (17.4)	**0.004**
CSF WBC>5	8/14 (57)	23/23 (100)	**<0.001**
ICU Admission	9/14 (64.3)	1/23 (4.3)	**<0.001**
Seropositive AIE N=33			
CCI>2	3/19 (15.8)	5/14 (35.7)	0.24
Anti-NMDAR	13/19 (68.4)	5/14 (35.7)	0.08
Anti-LGI1	3/19 (15.8)	4/14 (28.6)	0.42
AMS	16/19 (84)	7/14 (50)	**0.03**
Dysautonomia	7/19 (37)	0	**0.01**
Need for a 2nd Line	9/19 (47.4)	1/14 (7.1)	**0.02**
ICU Admission	9/19 (47.4)	1/14 (7.1)	**0.02**
Seronegative AIE N= 41			
CCI>2	10/24 (41.7)	7/17 (41.1)	0.98
AMS	17/24 (70.8)	10/17 (58.8)	0.42
Dysautonomia	1/24 (4.2)	1/17 (5.9)	1
Need for a 2nd line	1/21 (5)	2/15 (13.3)	0.56
ICU admission	10/24 (41.7)	5/17 (29.4)	0.52

Conclusion

- AIE demonstrated significantly higher healthcare utilization compared to HSE.
- There was no significant difference in healthcare costs nor outcomes at discharge between seronegative and seropositive AIE patients.
- Seropositive AIE patients more frequently received extensive inpatient immunotherapy, such as PLEX and second-line immunotherapy.
- Comorbidities, immunosuppression, altered mental status, focal neurological deficits, and absence of CSF pleocytosis, were associated with prolonged hospitalization in HSE.

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Mr Robin Münzing

Department of Neurology, University Hospital Ulm, Ulm, Germany

Intrathecal synthesis of target-specific IgG is more prominent in autoimmune encephalitides with NMDA, AMPA or GABAB receptor antibodies compared to those with CASPR2, DPPX, LGI1 and IgLON5 antibodies – a systematic review of the literature

After his high school diploma in 2017, Mr Robin Münzing joined the German armed forces and studied medicine at the University of Ulm. He obtained his medical degree in 2024 and started working in the Department of Internal Medicine of the Armed Forces Hospital Ulm. In May 2023, he started research work for his doctoral thesis under supervision of Prof. Jan Lewerenz in the Department of Neurology, University Hospital Ulm.

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Intrathecal synthesis of target-specific IgG is more prominent in autoimmune encephalitides with NMDA, AMPA or $GABA_B$ receptor antibodies compared to those with CASPR2, DPPX, LGI1 and IgLON5 antibodies - a systematic review of the literature

Robin Münzing and Jan Lewerenz

Department of Neurology, University Hospital Ulm, Ulm, Germany

Background & Introduction:
Autoimmune encephalitides (AIE) defined by the presence of neuronal surface antibodies of different subtypes represent a heterogenous group of diseases. Cerebrospinal fluid findings (CSF) in the different antibody-defined AIEs are also quite heterogenous, with common inflammatory changes in AIE with NMDA, AMPA and GABAB receptor antibodies but infrequent inflammatory CSF in those with CASPR2, LGI1 and IgLON5 antibodies (Blinder & Lewerenz, 2019). We asked whether those AIE subtypes with frequent CSF inflammation may also have more pronounced and/or frequent intrathecal synthesis (IS) of target-specific IgG as compared to those with less frequent CSF inflammation.

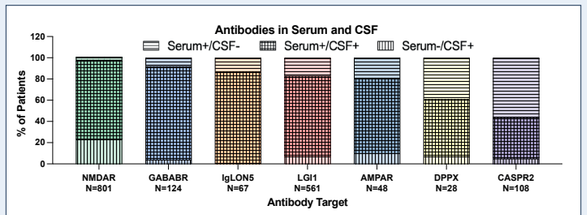

Figure 1: The antineuronal antibodies are detectable at different proportions in CSF only, CSF and serum, and serum only in the seven most common autoimmune encephalitis subtypes. The percentage of patients with antibodies in serum only, both serum and CSF and CSF only is depicted as indicated.

Methods:
A comprehensive literature search of papers published from January 1st 2019 to December 31st, 2023 was performed using the PubMed database. Publications analyzed whether individual patients or cohorts with AIE with NMDAR, AMPAR, $GABA_BR$, DPPX, CASPR2, LGI1, IgLON5, $GABA_AR$ or GlyR antibodies were described with the results of antibody testing in CSF and serum reported. For all patients, it was analyzed whether antibodies were reported positive in CSF and serum, serum only or CSF only. For those with antibody titers in CSF and serum reported, the CSF/serum antibody ratio was calculated. For patients with negative serum but CSF titer available, the serum titer was set to 1, for those with negative CSF, the CSF titer was set to 0.1. A CSF/serum ratio of >0.01 was rated as proof of target-specific IS, a ratio of <0.001 as absence of IS, while a ratio of 0.01 to 0.001 was rated as possible IS.

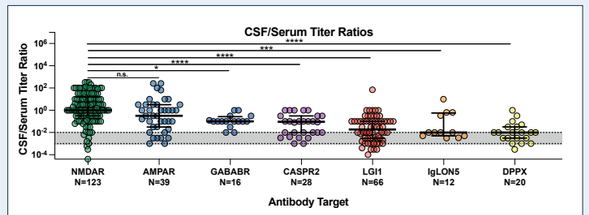

Figure 2: The CSF/serum titer ratio of antineuronal antibodies varies widely among different autoimmune encephalitis subtypes although intrathecal synthesis occurs in all. Depicted is the median and interquartile range. Grey area: CSF/serum titer ratio 0.01 - 0.001, possible intrathecal synthesis, above: definite intrathecal synthesis. Statistical comparison by Kruskal-Wallis test with pairwise posthoc Dunn's test as indicated.

Results

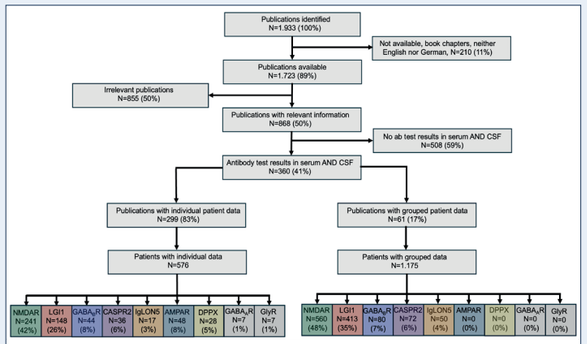

Table 1: Results of the literature search. Individual data: data describing single patients, grouped data: data describing cohort of patients. Due to the low number of CSF and serum data, $GABA_AR$ and GlyR antibodies were analyzed further

	NMDAR	LGI1	$GABA_BR$	CASPR2	IgLON5	AMPAR	DPPX	$GABA_AR$	GlyR
Total, N (% of all)	801 (46)	561 (32)	124 (7)	108 (6)	67 (4)	48 (3)	28 (2)	7 (<1)	7 (<1)
Sex known, N (%)	632 (79)	516 (92)	124 (100)	100 (93)	16 (24)	48 (100)	28 (100)	7 (100)	7 (100)
Female/Male, N (%)	405/227 (64/36)	204/312 (40/60)	43/81 (35/65)	35/65 (35/65)	9/7 (56/44)	21/27 (44/56)	11/17 (39/61)	3/4 (43/57)	2/5 (29/71)
Age known, N (%)	241 (30)	147 (26)	44 (35)	36 (33)	17 (25)	48 (100)	28 (100)	7 (100)	7 (100)
Age (years), Median (IQR)	23 (15-32)	60 (48-69)	62 (53-67)	52 (10-67)	65 (61-74)	51 (30-63)	52 (32-61)	54 (44-65)	60 (41-73)
CSF/Serum Titer known, N (%)	123 (15)	66 (12)	16 (13)	28 (26)	12 (18)	39 (81)	20 (71)	2 (29)	1 (14)

Table 2: Basic demographic characteristics. Demographic characteristics of the combined cohort of patients with the antibody-defined autoimmune encephalitis and the proportion known with CSF and serum antibody titers

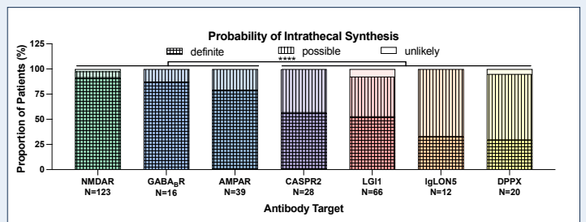

Figure 3: The frequency of definite intrathecal synthesis is higher in patients NMDAR, $GABA_BR$, and AMPAR antibodies compared to those CASPR2, LGI1, IgLON5, and DPPX antibodies. Definite, possible, and unlikely intrathecal synthesis (IS) was categorized as in Figure 2 and shown as percentage of patients in each category per autoimmune encephalitis subtype. Frequency of definite IS was compared to possible/unlikely IS in subtypes grouped as indicated using a Fisher's exact test.

Conclusion

Our systematic analysis of published data indicates that

- Seropositivity and detection of antibodies in CSF differs among the seven most common autoimmune encephalitis subtypes
- Detection of antibodies in CSF only is most common in autoimmune encephalitis with NMDAR antibodies
- Negative antibodies in serum are most frequent for CASPR2 and DPPX
- CSF/serum titer ratios indicate that the extent of intrathecal synthesis of antineuronal antibodies differs substantially among different autoimmune encephalitis subtypes with definite intrathecal synthesis being most frequent in patients with NMDAR, $GABA_BR$ and AMPAR antibodies
- However, our data indicate that intrathecal synthesis of disease specific antibodies occurs in all autoimmune encephalitis subtypes analyzed
- Thus, intrathecal synthesis of disease-specific antibodies might be pathophysiological relevant in most if not all autoimmune encephalitis subtypes

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Sarah A Boardman

University of Liverpool, Faculty of Health & Life Science, Institute of Infection Veterinary & Ecological Sciences, Department of Clinical Infection, Microbiology and Immunology, UK

Investigating direct and indirect effects of viral infections on the blood-brain barrier

Sarah is a PhD student at the University of Liverpool and is a member of the Infection Neuroscience Laboratory directed by Professor Benedict D. Michael. Her research focuses on using in vitro modelling, specifically microfluidic chips, to model the blood brain barrier. Using this model, she aims to look at blood brain barrier damage and the effect on the wider neurovascular by infecting the model with HSV-1 and VZV. As well as inhibiting key chemoattractant protein pathways to assess if any are suitable for future work to develop therapeutics that prevent infections causing damage to the brain.

Investigating direct and indirect effects of viral infections on the blood-brain barrier

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INTRODUCTION

Viral infections can cause severe neurological complications, such as encephalitis and seizures[1]. The brain's blood-brain barrier (BBB) acts as a shield against toxins and microorganisms. Key BBB components—endothelial cells, basement membrane, pericytes, and astrocytes —maintain their selective permeability through tight and adherens junctions between endothelial cells[2] (Figure 1).

Viruses can compromise the BBB, increasing permeability and allowing pathogens and immune cells to enter, triggering inflammation. The mechanisms of BBB crossing by pathogens, either directly affecting brain cells or indirectly through endothelial infection, remain under investigation and thus limit treatment options[3].
Animal models, though insightful, pose ethical and cost challenges, while *in vitro* models like transwell assays lack physiological shear stress. Microfluidic "brain-on-a-chip" devices better simulate the BBB environment and offer a promising alternative[4].

This project will use microfluidic chips to study how Herpes simplex virus-1 (HSV-1) and Varicella zoster virus (VZV) impact the BBB. By examining viral-induced BBB damage and testing inhibitors of chemoattractant pathways, we aim to uncover potential therapeutic targets for viral neurological diseases.

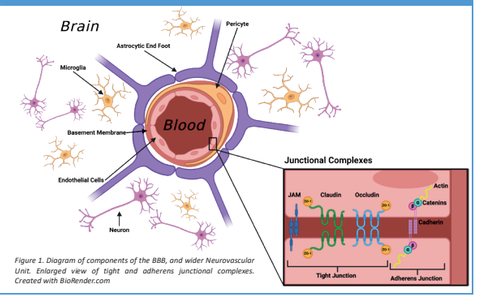

Figure 1. Diagram of components of the BBB, and wider Neurovascular Unit. Enlarged view of tight and adherens junctional complexes. Created with BioRender.com

MICROFLUIDIC CHIP MODEL

Microfluidic chips provide a promising alternative to animal models for studying BBB interactions. This project will use these chips to create a "brain-on-a-chip" by co-culturing human cerebral microvascular endothelial cells (CMECs) and primary human astrocytes (Figure 2). The chip design enables chemical and cellular communication between these cells, mimicking *in vivo* conditions.

Benefits of microfluidic chips:

- Accurately models shear stress and perfusion via syringe pump
- Maintains spatial separation of cell types, reflecting *in vivo* physiology
- Optimizes culture conditions for each cell type
- More closely replicates infection and drug effects seen in living systems

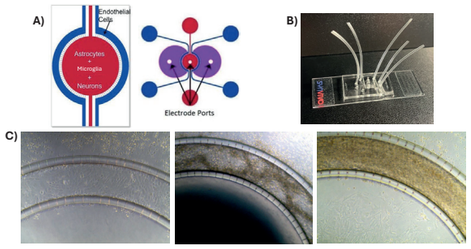

Figure 2. A) Schematics of microfluidic devices. Apical chamber (blue) are for culture of vascular cells, while basolateral chamber (red) are for culture of brain tissue cells. Micro-channels enable communication of cells between chambers, B) Image of microfluidic chip, C) Microscopy image of hCMEC/D3 cultured in apical channel on day 4, day 7 and day 11. Figure 2A taken from Synvivo website: https://www.synvivobio.com/synbbb/

TRANSENDOTHELIAL ELECTRICAL RESISTANCE (TEER) OVER TIME

TEER measurements quantitatively assess endothelial monolayer integrity and permeability[5]. Electrodes placed on either side of the monolayer measure electrical resistance using an analyser that applies a voltage and records the resulting current (Figure 3). High impedance (kΩ) indicates an intact monolayer, while decreases show an increased permeability.

Figure 4 shows improved monolayer integrity from day 0 (seeding) to day 12, with flow introduced on day 3. Shear stress enhances monolayer integrity, reducing permeability.

In transwell models, a TEER value ≥50 kΩ signifies an intact monolayer[6]. These data show values exceeding 50 kΩ by day 4, with some channels reaching nearly 400 kΩ by day 12. The introduction of other cell types (e.g. astrocytes) is likely to further improve resistance.

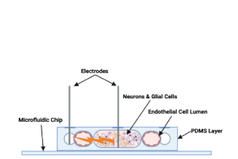

Figure 3. Diagram of microfluidic chip with electrode placement for measuring TEER. Created with BioRender.com

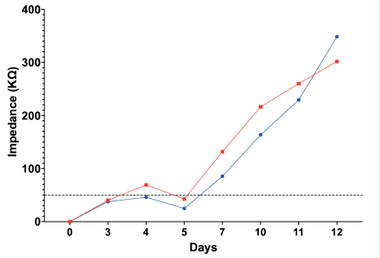

Figure 4. TEER measurements taken over a period of 12 days. Each line represents an individual channel where hCMEC/D3 have been cultured. Black dotted line represents 50KΩ.

IMMUNOFLUORESCENT STAINING

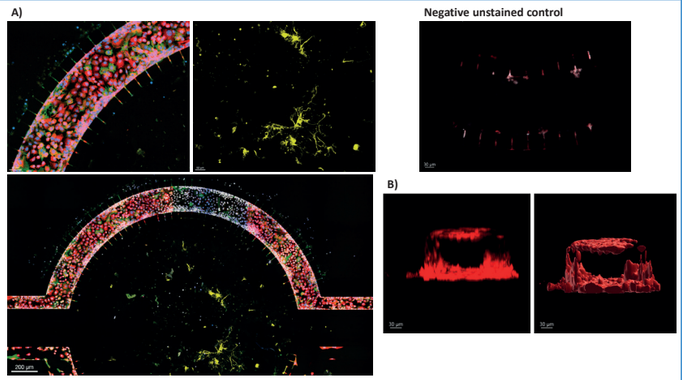

Figure 5. Expression of the tight junction protein ZO-1 (red), endothelial cell marker CD31 (green), astrocytic marker GFAP (yellow) and DAPI (nuclei, blue) in hCMEC/D3 and primary human astrocytes. A) Whole apical channel showing high expression of ZO-1, as well as CD31 from endothelial cells and GFAP expression from astrocytes – close up view displayed above whole channel as well as negative control, B) Cross-sectional view of endothelial cell forming an inner lumen in apical chamber (Left: Maximum Intensity Projection view; Right: 3D computer rendered modelled surface).

To verify tight junction expression, ZO-1, proteins were visualized via immunofluorescent staining and fluorescent microscopy. The high amount of tight junction protein expression confirms the formation of a robust endothelial barrier in the apical chamber. Figure 5A shows ZO-1 (red) expression across the entire population of cells and also expression of CD31 (green), an endothelial cell marker. Additionally, detection of the astrocytic marker, GFAP (yellow), was observed from primary human astrocyte cultured in the central chamber.

Figure 5B illustrates the growth of a lumen structure, replicating the formation of a blood vessel.

VIRAL INFECTION IN BBB MODEL

This project will investigate two Herpesviridae viruses with differing mechanisms of neurological impact:

HSV-1: Direct, neural infection
VZV: Indirect, endothelial infection

TEER was used to assess endothelial monolayer integrity over 24 hours post-infection (PI) with HSV-1, Readings were taken pre-infection, every 2 hours for 8 hours PI, and at 24 hours PI. Supernatant from CMECs were collected at each time point for Luminex analysis to assess changes in chemoattractant protein profiles. Luminex panels are based on prior INL research identifying key cytokines, chemokines, and brain-injury markers.

Preliminary data from HSV-1 infection in a CMECs and primary human astrocyte co-culture didn't reveal a clear impact on TEER (Figure 6), possibly due to low astrocyte density in the central chamber or too few viral particles. Impedance changes are presented as percentage change, with 100% representing pre-infection values.

To assess whether the MOIs previously used can cause an effect, future plans are to use a Maestro impedance 96-well plate to culture CMECs and astrocytes, measure impedance non-invasively, and monitor changes over 48 hours PI.

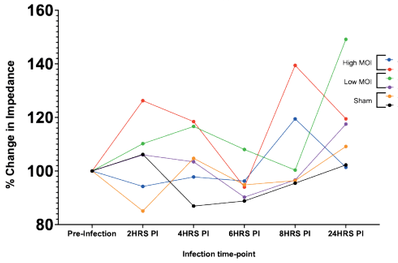

Figure 6. Percentage change in impedance over a period of 24hrs. Channels cultured with hCMEC/D3 had their TEER measured prior to infection with HSV-1 and then every 2hrs post-infection (PI) for 8hrs and a final measurement at 24hrs. Astrocytes in the central chamber were infected with either an MOI of 0.5 (blue and red); MOI of 0.05 (green and purple); or sham infected with Vero cell supernatant (orange and black).

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NIHR Health Protection Research Unit in Emerging and Zoonotic Infections at University of Liverpool

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Simona Serra, PhD student

Department of Neuroscience, Karolinska Institutet, Stockholm, Sweden

Broad-spectrum antimicrobial activity of TKD-008A and TKD-008B: A new antimicrobial drug with neuroprotective properties

Simona Serra, PhD student received her B.Sc. with honours in Biotechnology from the University of Salento (Lecce, Italy) in 2021, followed by an M.Sc. with honours in Molecular and Cellular Biology from the University of Bologna (Bologna, Italy) in 2023.
Her undergraduate studies were supported by several scholarships, including an Erasmus grant from the University of Bologna and a Stifelsen Dementia Association scholarship. Simona is now a doctoral student in F. Iovino's laboratory at the Department of Neuroscience, Karolinska Institutet (Stockholm, Sweden), where she investigates 1) the molecular mechanisms of neuronal damage during the pathogenesis of meningoencephalitis, primarily caused by Streptococcus pneumoniae (pneumococcus), and 2) the antimicrobial properties of specific human peptides that can be used as new neuroprotective treatments against bacterial meningoencephalitis. Her latest study, published in iScience, demonstrates how a single amino acid substitution significantly enhances the cytotoxicity of pneumolysin (the main pneumococcal toxin) towards neuronal cells (DOI: 10.1016/j.isci.2024.109583).

Broad-spectrum antimicrobial activity of TKD-008A and TKD-008B: A new antimicrobial drug with neuroprotective properties

Simona Serra, Federico Iovino
Department of Neuroscience, Karolinska Institutet, Stockholm, Sweden.

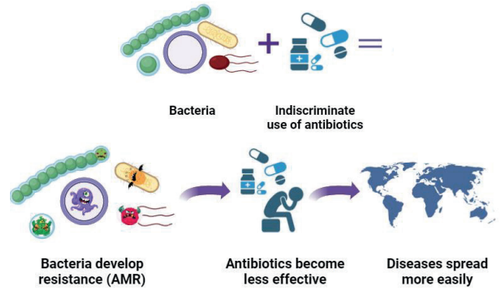
Conclusions

TKD-008A and its active peptide TKD-008B show antimicrobial effect against both Gram-positive and Gram-negative bacteria. This breakthrough discovery has the potential to pave the way for a novel antibiotic therapy.

CFU count, OD measurement, Immuno-Electron Microscopy and LDH assay display a notably significant bactericidal effect of TKD-008A and TKD-008B peptide on bacteria, coupled with no cytotoxicity towards neuronal cells.

Introduction

- Antimicrobial resistance (AMR) is increasing at an alarming rate.
- AMR is due to an inappropriate use of antibiotics



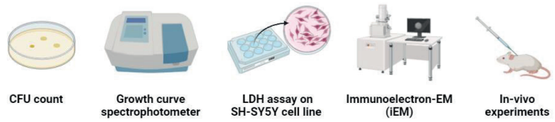
Aim of the study

TKD-008A and TKD-008B peptide show a great **antimicrobial effect**, overcoming the current AMR burden. This research has the potential to introduce a new antibiotic drug into clinical use, thereby improving health outcomes related to infections and reducing the ease with which the bacteria can spread.

Methods

The bacteria involved in this study are:

- Gram-positive → *Streptococcus pneumoniae*
- Gram-negative→ *Escherichia coli* and *Pseudomonas aeruginosa*



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More details:

Results

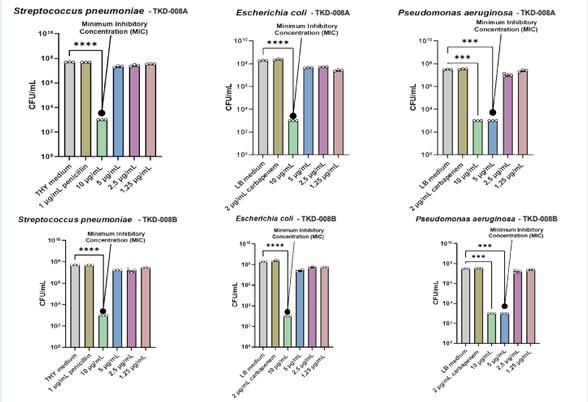

Fig 1. Determination of the **Minimum Inhibitory Concentration (MIC)** of TKD-008A and TKD-008B. Bacterial strains tested: penicillin-resistant *Streptococcus pneumoniae* SP6, carbapenem-resistant *Escherichia coli* ST131 and carbapenem-resistant *Pseudomonas aeruginosa* P476.

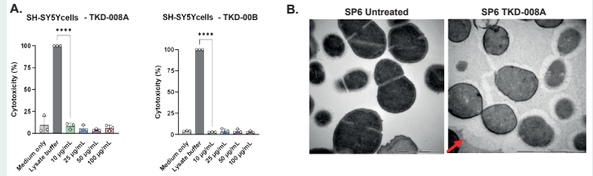

Fig.2 A) Cytotoxicity assessment of TKD-008A and TKD-008B on SH-SY5Y-derived human neuronal cells. **B) Cryo-electron microscopy** analysis of *Streptococcus pneumoniae* SP6: untreated and after treatment with TKD-008A .

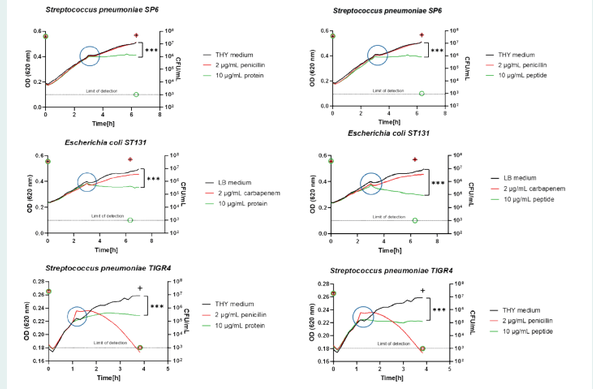

Fig 3. Assessment of bacterial growth and bacterial viability through CFU count and OD measurement. Bacteria were challenged with MIC concentrations of TKD-008A, TKD-008B, penicillin and carbapenem.

Ongoing work:

- iEM tomography
- In-vivo experiments
- Mechanism of action

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Arboviral encephalitis in Pakistan 2017-2020 – clinical and laboratory features and neurological outcomes

Dr Sobia Khan is a senior postgraduate trainee (clinical fellowship) in medical microbiology at the Aga Khan University Hospital. She has represented her institute at multiple national and international research forums and has received an intramural grant for work on arboviral surveillance, specially focusing insecticide resistance. Her fellowship dissertation is focused on acute meningoencephalitis in Pakistan.

Arboviral encephalitis in Pakistan 2017-2020 – Clinical and laboratory features and neurological outcomes

Sobia Khan, Tazeen Fatima, Erum Khan, Bushra Jamil, and Sadia Shakoor

INTRODUCTION

Neuroinvasive arboviruses include flaviviruses i.e. West Nile Virus (WNV), Dengue virus (DENV), tick-borne encephalitis virus (TENV), Zika virus (ZIKV), and Japanese encephalitis virus (JEV) and alphaviruses like Chikungunya virus (CHIKV), Eastern Equine Encephalitis virus (EEEV), Western Equine Encephalitis virus (WEEV), Venezuelan Equine Encephalitis virus (VEEV) that are known to be endemic and cause recurrent outbreaks in Pakistan. Owing to non-specific clinical features mimicking acute febrile illness, early recognition of these infections is essential for decreasing long term disability and control through improved surveillance.

OBJECTIVES

- To assess frequency of arboviral encephalitis in hospitalized cohort at a tertiary care center in Karachi, Pakistan.
- To assess laboratory parameters and clinical features in patients with flaviviral encephalitis.

MATERIALS AND METHODS

From 2017 to 2020, medical records of all patients with acute encephalitic syndrome (AES) admitted to a tertiary care center in Karachi were reviewed.

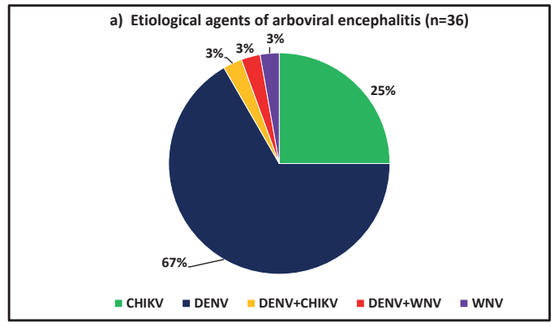
AES was defined as altered mental status lasting >1 day with minor criteria per Venkatesan et al (2013).

Flaviviral and CHIKV encephalitis were diagnosed by excluding other etiologies and presence of flaviviral IgM or chikungunya IgM in acute sera from a larger cohort of patients analyzed by Shakoor S. et al. [4]

Duration of illness before presentation, symptoms, neurological signs, radiological and EEG abnormalities, duration of hospitalization, hospital mortality and Rankin score at discharge were noted. Data was analyzed in Prism version 10.

RESULTS

- 522/668 patients could be attributed to possible infectious etiology.
- 36/522 episodes of AES (6.9%; 95% CI=48.8-94.2) were confirmed to be due to flaviviruses.
- Of these; majority i.e. 24 (66.6%) were positive for DENV IgM. (Fig 1a)
- Median age of patients was 40.5 years (IQR 25-66), 61% were male.
- Median CSF leukocyte counts were 24 cells/µl (IQR 5.5-72).
- Median symptom duration at presentation was 5 days (IQR 3-7).
- Clinical features included fever in all patients, seizures in 14 (38.9%, 95%CI 23-56.5), and focal neurological signs in 6 (6.7%, 95%CI 6.4-32.8), and slurred speech or aphasia in 4 (11.1%, 95%CI 3.1-26). (Fig 1b)
- EEG was performed in 21 patients, with abnormalities detected in all patients.
- Median duration of hospitalization in arboviral AES patients was 7 days (IQR 4-10.5).
- Five (n=5, 13.9%, 95%CI 4.7-29.5) patients died (4 flaviviral AES, one chikungunya AES), one patient (chikungunya AES) was discharged with a Rankin score of 5.
- Clinical features, CSF leukocytosis or EEG abnormalities did not predict death or higher disability scores on discharge.



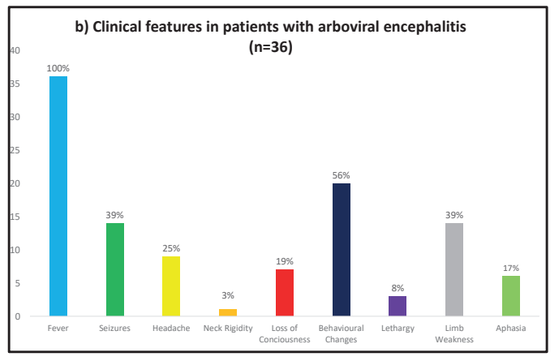


Fig 1: (a) Etiological agents of flaviviral encephalitis.
(b) Clinical features in patients with flaviviral encephalitis.

Discussion

Our study shows that arboviruses are an important cause of AES in Pakistan contributing up to 7% of cases, majority i.e. 66.6% attributed to DENV. Median duration to development of symptoms was 5 days which is in concordance with other studies.[1]

In South Asia, DENV is endemic in almost all countries, JEV transmission is widespread, and limited WNV transmission has been reported, most frequently from India.[2] In Pakistan, no definitive reports of Zika virus infection have been identified to date; however, Butt et al estimated the potential of introduction of Zika virus through international travel and that the highest proportion of high-risk travelers is from Singapore.[3] Similarly, preliminary evidence of JEV transmission exists, however, confirmation was not possible due to flaviviral serological cross reactivity and other factors.[2]

Thus, our study highlights the need of enhanced vector surveillance and arboviral control programs for early detection of potential outbreak in this part of the world.

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Epidemiology and clinical characteristics of varicella zoster virus reactivation encephalitis among patient with acute encephalitis syndrome in Southern Pakistan

Dr Sobia Khan is a senior postgraduate trainee (clinical fellowship) in medical microbiology at the Aga Khan University Hospital. She has represented her institute at multiple national and international research forums and has received an intramural grant for work on arboviral surveillance, specially focusing insecticide resistance. Her fellowship dissertation is focused on acute meningoencephalitis in Pakistan.

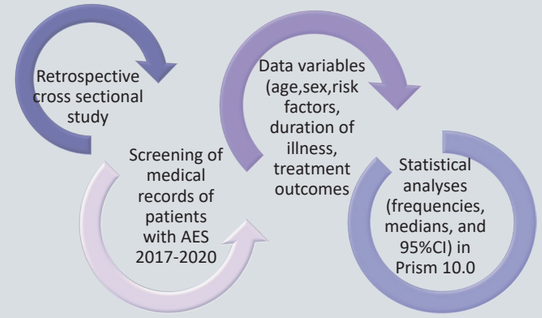
Epidemiology and clinical characteristics of Varicella Zoster Virus reactivation encephalitis among patients with acute encephalitis syndrome in southern Pakistan

Sobia Khan, Erum Khan, Bushra Jamil, Asima Shahid, Najma Shaheen, Sadia Shakoor

Introduction

Varicella Zoster Virus (VZV) is a highly contagious neurotropic virus causing a reactivation illness (herpes zoster- HZ) in immunocompromised or elderly individuals. Central nervous system (CNS) infection in HZ presents with various CNS pathologies including vasculitis, myelopathy, meningitis, and encephalitis. VZV encephalitis testing is not widely available in Pakistan and is often not performed leading to an under recognized illness burden and lack of therapeutic or preventive measures that result in high morbidity. We present 11 cases of VZV encephalitis from a larger cohort of patients with infectious meningitis analyzed by Shakoor S. et al[5] to describe its burden, clinical characteristics, and outcomes of infection in Pakistan.

Methods



Results

- Of 520 patients, 11 had VZV encephalitis confirmed by a multiplex syndromic PCR test (Filmarray Meningitis/Encephalitis, Biomereiux, USA).
- Patients had a median age of 50 (IQR45-63) years.
- Male: female ratio was 4.5.
- All patients were immunocompetent; 27% were diabetic.
- Patients presented after a median of 2 (IQR 2-8) days of illness onset.
- Clinical features included fever and altered consciousness (in 100%), generalized seizures in 18%, neck rigidity in 9%, focal signs in 18%, Bell's palsy in 9%, and a rash consistent with zoster in 36.4%.
- Electroencephalographic abnormalities (theta delta slowing) were observed in 18%.
- Imaging abnormalities were seen in 66.6% (6 of 9) patients, and 3 of 9 (33.3%) had evidence of vasculitis and infarcts.
- All patients received IV acyclovir.
- The median duration of hospitalization was 4 (IQR3-11) days.
- 36% of patients were discharged home in a persistent vegetative state, despite adequate treatment with acyclovir.

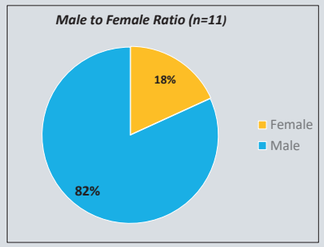


Figure 1. Male to female ratio of patients with VZV encephalitis.

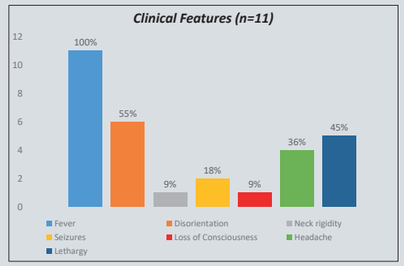


Figure 2. Clinical features of patients with VZV encephalitis.

Discussion

Similar to Mirouse, A et al [1], our study concluded that VZV encephalitis is more frequent in middle aged men and fever is most common clinical manifestation. However, in contrast to our findings, 2/3rd of the patients in previously conducted studies presented with rash and around 40-80% of them had an underlying immunocompromised condition.[1,2] Nevertheless, a number of case reports have shown its occurrence in immunocompetent adults as well.[3,4]

Conclusion

VZV encephalitis incurs a significant burden and morbidity among middle aged immunocompetent Pakistanis. Routine inclusion of VZV in syndromic meningoencephalitis test panels in Pakistan can improve detection. High morbidity among adults may be prevented by regular varicella vaccination in childhood and zoster vaccine in diabetic adults.

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ENCEPHALITIS CONFERENCE 2024

Ms Suzanne Franken

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The role of IgG subclasses in CASPR2 Autoimmune Encephalitis

Ms Suzanne Franken is a PhD candidate at the autoimmune encephalitis research group of the Erasmus MC University Medical Center in Rotterdam. She obtained her Masters in Biomolecular Sciences at the Vrije Universiteit of Amsterdam in 2020. During her Masters internship, she studied the presence of anti-glycolipid antibodies in neuralgic amyotrophy. Before starting her PhD, she worked on various projects regarding IgG subclasses in CIDP and IgLON5 autoimmune encephalitis.

The role of IgG subclasses in CASPR2 Autoimmune Encephalitis

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S.C. Franken[1], T. Brand[1], M.R. Vermeieren[2], M.M. van Duijn[1], J.M. de Vries[1], M.J. Titulaer[1]

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Introduction

Though anti-CASPR2 encephalitis is among the more **frequently** identified AE subtypes and many patients' relapse, studies regarding **long-term outcomes and relapse risk** are scarce and unknown. Since IgG1 and IgG4 have different effector functions, it has been suggested that **IgG1/IgG4** frequency relevant for both. We examined the presence of IgG subclasses in sera and if they relate to long-term outcome.

Methods

A total of **44 Dutch CASPR2 AE patients** of whom serum and/or CSF tested positive by both IHC and CBA between 2007 and 2024 were included. A method for **high-throughput screening** for anti-CASPR2 antibodies was developed and optimized, combining the *in-house* CBA and the Opera Phenix High Content Screening System. Using this method, sera of **25/44** patients were tested for anti-CASPR2 **IgG1 and IgG4** antibodies.

Conclusions

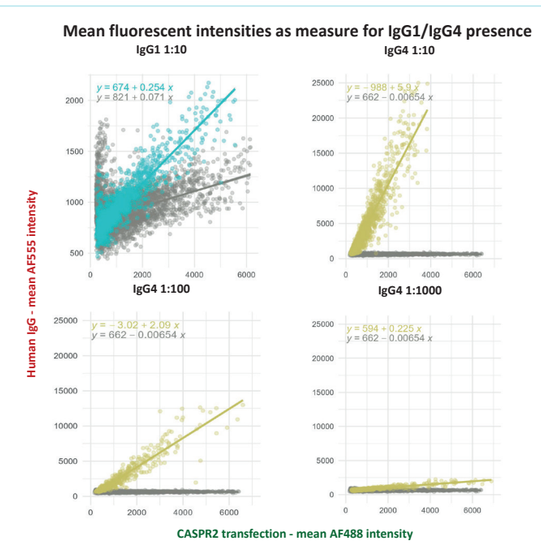
Combining CBA with the Opera Phenix System enables for high-throughput, quantitative assessment for antibodies. Almost all patients harbour CASPR2 IgG4 antibodies, while ~30% present IgG1, albeit often in lower quantities. Patients who have CASPR2 IgG1 antibodies show a more severe clinical phenotype, but no other differences were identified in this preliminary data.

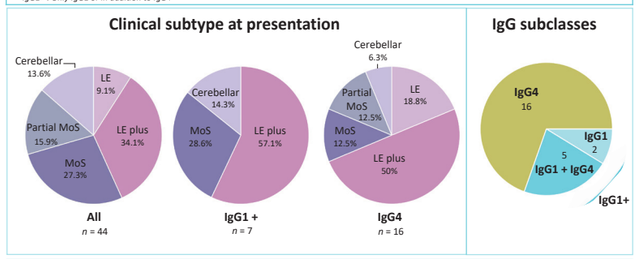
Results

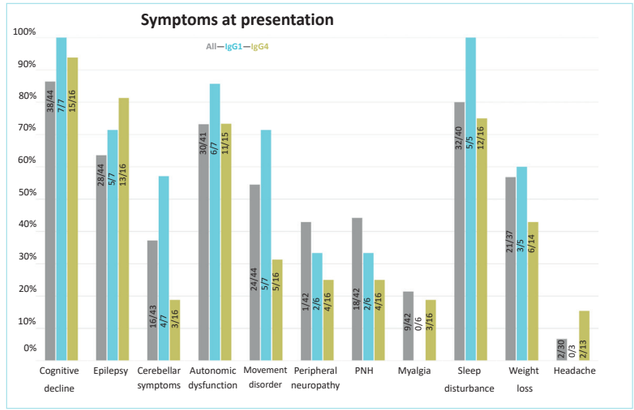
Table I: Demographic characteristics CASPR2 AE patients.

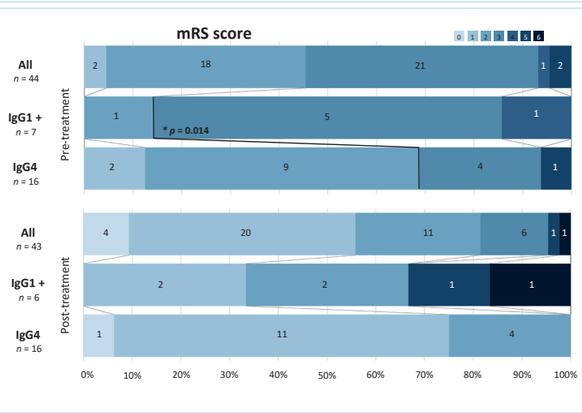
Characteristics	All	IgG1+	IgG4	*p*-value
	n = 44	*n* = 7	*n* = 16	
Male sex, n (%)	42 (96)	7 (100)	16 (100)	
Age, y, median (range; IQR)	66 (46; 60.3-71)	64 (18; 60-68)	67 (25; 62-96.5)	0.44
Diagnostic delay, m, median (range; IQR) (*n* = 42)	8 (105; 3.8-17.3)	6.5 (18; 2.5-13.8)	8 (45; 4-19.3)	0.33
Follow-up time, m, median (range; IQR) (*n* = 43)	55 (192; 35-78)	63 (83; 28.8-84)	52 (78; 15.8-72.5)	0.49
Relapse, n (%)	24/44 (55)	3/7 (43)	12/16 (75)	0.18
Tumour at presentation, n (%)	12 (27)	2 (29)	3 (19)	0.62
MRI FLAIR/T2 abnormalities, n (%)	16/41 (39)	3/6 (50)	9/15 (60)	1.00
RIA, median (range; IQR) (*n* = 41)	394 (754; 296-477)	414 (395; 365-422)	385 (754; 260.5-480.5)	0.67

IgG1+ : Only IgG1 or in addition to IgG4









Abbreviations

AE: Autoimmune Encephalitis | CBA: Cell-Based Assay | CASPR2: Contactin-associated protein-like 2 | LE: Limbic Encephalitis | MoS: Morvan Syndrome | mRS: Modified Ranking Scale | PNH: Peripheral Nerve Hyperexcitability | RIA: Radioimmunoassay

Funding

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Single centre experience of paediatric myelin oligodendrocyte glycoprotein (MOG) antibody associated encephalitis

Miss Shruthi Mundasad, works as Paediatric Neurology registrar at Kings College Hospital. She has completed Primary medical school and Paediatric training in India, completed MRCPCH at London.

Single centre experience of paediatric Myelin oligodendrocyte glycoprotein (MOG) antibody associated encephalitis

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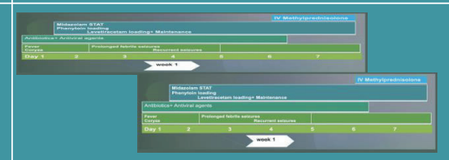
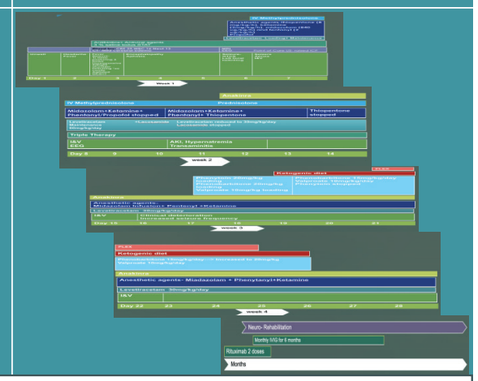
Introduction:
Myelin oligodendrocyte glycoprotein (MOG) antibody-associated disease (MOGAD) is an inflammatory demyelinating disease of the central nervous system with the presence of conformation-sensitive antibodies against MOG.
Whilst it was first associated with demyelination, more recently the phenotypic spectrum has expanded to include MOG associated encephalitis.

Aims and Objective:
To describe the clinical spectrum and outcomes of MOG-Ab associated encephalitis in paediatric patients to highlight the features and challenges of this condition. We reviewed series of children with MOG-Ab-associated encephalitis, manifesting with a distinctive neurological illness, imaging pattern and clinical outcomes.

Case	EEG/Neuroimaging	Treatment
1)11mon/Male Prolonged febrile seizure, encephalopathy and hyperkinetic movement disorder Lab- CSFWBC <5/c mm MOG Ab Blood- negative,CSF- positive	EEG-Focal seizures arising from midline and left cento temporal regions spreading to left posterior regions MRI- restricted diffusion frontal and extending into parietal white matter, sparing the peri-rolandic region Repeat imaging after steroids – signal change resolved with normal basal ganglia.	
2)13mon/Male Short history of fever and upper respiratory tract symptoms followed by left focal motor seizure with secondar y generalization evolving into status Lab- CSF WBC<5/cmm MOG Ab Blood-negative, CSF-positive	EEG- slow with left focal epileptiform discharges MRI - widespread bilateral T2 high signal and restricted diffusion with predominance of the posterior cor tex and relative subcortical sparing, together with involvement of the deep grey matter, not in keeping with ADEM.	
3)10yr/Male Background of CF,one week history of being unwell, evolved into headaches, vomiting, blurring of vision, confusion and stiffness of all limbs CSF WBC 9/cmm MOG Ab Blood-Positive, CSF- Negative	EEG-Status epilepticus CT – raised ICP features, MRI - ongoing swelling with some patchy signal in thalami and basal ganglia	
4)12yr/Female Headache, seizures, encephalopathy CSF WBC<5/cmm MOG Ab Blood-Positive, CSF- Negative	EEG-Markedly abnormal record containing frequent and multiple types of clinical and electrographic seizures MRI- cortical changes and diffuse leptomeningeal enhancement, some of the cortical signal abnormality and restricted diffusion attributed to seizures	

Conclusions:
Anti-MOG-associated cortical encephalitis has a wide spectrum of phenotypical presentation and is potentially devastating. Common symptoms include seizures, headache, encephalitic features, and cortical symptoms, such as paresis. MOG antibody testing is infrequently included in the diagnostic work-up of patients with suspected autoimmune encephalitis. Children with MOGAD present with diverse imaging patterns which correlate with the main demyelinating phenotypes as well as age at presentation.
Paediatric MOGAD patients typically respond rapidly to IVMP in the acute attack. IVIG and PLEX are second-line treatment options during acute attack. Early recognition and treatment may be essential to optimise outcomes.

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ENCEPHALITIS CONFERENCE 2024

Dr Thomas Agerbo Gaist

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Autoimmune Encephalitis in Denmark: Results from the DanAIMS study

Dr Thomas Gaist is a physician and PhD research fellow studying autoimmune encephalitis under the supervision of Professor Morten Blaabjerg at the Neurological Research Unit at Odense University Hospital, Denmark. The focus of the PhD is to provide a comprehensive overview of the epidemiology and long-term outcomes of AE in Denmark. By establishing a nationwide AE database containing all AE patients identified in Denmark from 2009-2023, national epidemiological trends can be studied. Using the database data in conjunction with the Danish national registries, the long-term socio-economic outcomes can be studied in detail. A national clinical study investigates the long-term cognitive and neuroimaging outcomes of the cohort.

Autoimmune Encephalitis in Denmark: Results from the DanAIMS study

Thomas Agerbo Gaist[1,2,3], Mette Scheller Nissen[1,2,3], Anna Christine Nilsson[2,3,4], Silke Funch[3], Charlotte Aaberg Poulsen[5], Klaus Hansen[6], Morten Blaabjerg[1,2,3]

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BACKGROUND

Awareness of autoimmune encephalitis (AE) has increased in recent years, yet basic information on the epidemiology of this rare disorder is scarce. Denmark, with its centralized antibody testing, free access to health care, and a plethora of medical and administrative registries available for research purposes, provides an ideal framework for studies of AE epidemiology. We therefore collected data on all patients with a verified sero-positive AE diagnosis in Denmark. We used this information to calculate basic epidemiologic measures of the occurrence of AE in Denmark over time.

METHODS

Utilizing data from a national Danish test centre, we identified all patients with positive for serum and/or CSF IgG neuronal surface antibodies (NSAb), GAD65 and GFAP in 2009-2023. We then verified the diagnosis of AE based on medical records. Patients with a verified AE-diagnosis were included in the cohort. We also included patients with non-encephalitic phenotypes that corresponded with the identified autoantibody (e.g., GAD65 positive Stiff-person spectrum disorder). Based on DanAIMS data and population census data, we calculated the crude incidence rate (IR) as the number of cases per one million person-years and analyzed temporal trends in incidence and antibody testing using linear regressions.

RESULTS

Figure 1: Distribution of patients by antibodies and cohort by sex

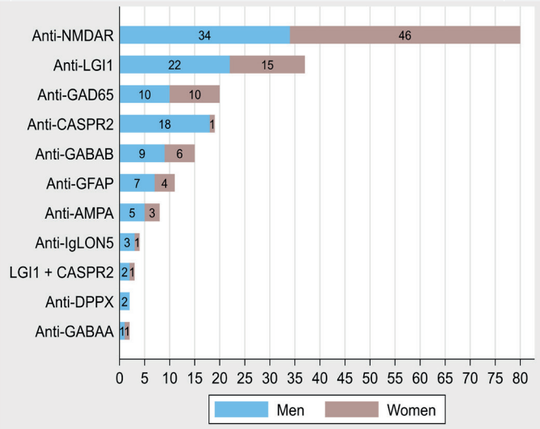


Figure 2: Patient characteristics by phenotype

	Full cohort	NMDAR encephalitis	Limbic encephalitis	Other encephalitis	Non-encephalitic
	N = 201	n = 76	n = 83	n = 24	n = 18
Age at diagnosis, median (range)	57 (3 - 88)	27 (3 - 81)	68 (9 - 88)	61 (4 - 81)	48 (18 - 77)
Sex, %female	44	59	35	25	44
Underlying cancer, %	17	12	27	4	11
Relapse, %	15	11	24	12	0
Death from AE or related cancer, %	13	7	20	21	0
Days from symptom onset to diagnosis, median (IQR)	41 (148)	26 (29)	84 (173)	30 (311)	270 (263)

Figure 3: Incidence of sero-positive AE and AE antibody testing in Denmark 2009-2023

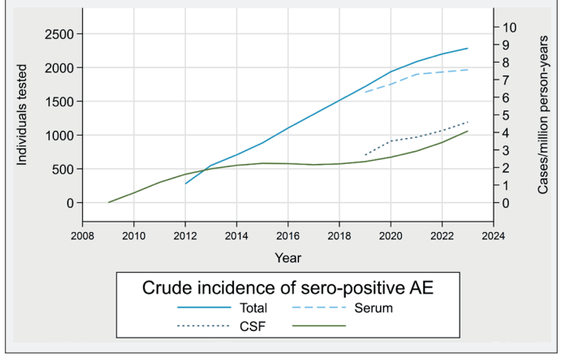
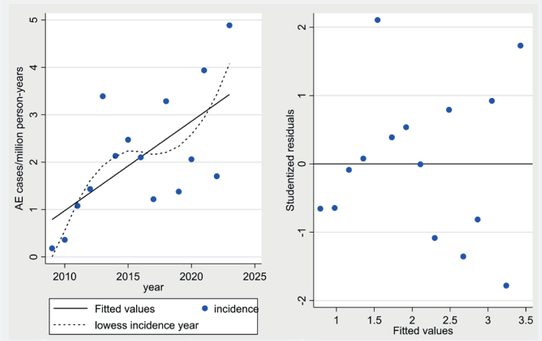


Figure 4: Linear regression of incidence over time and studentized residuals



CONCLUSIONS

The crude incidence of sero-positive AE in Denmark is comparable to those found in previous studies from other countries. From 2009-2023, the incidence has increased significantly, which indicates that not all Danish AE patients are receiving the correct diagnosis. The number of individuals tested for AE over time is not significantly associated with the incidence, indicating that clinician awareness in Denmark is increasing.

SDU University of Southern Denmark

DANISH REFERENCE CENTER FOR AUTOIMMUNE ENCEPHALITIS AND RELATED DISORDERS

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Single cell immune cell survey identifies a pathogenic role for T cells in anti-NMDA receptor encephalitis

Dr Thomas Johnson is a Foundation Year 1 Doctor undertaking an Academic Specialised Foundation Programme in Oxfordshire. Alongside his clinical training at Oxford University Hospitals, he is undertaking research at the Wellcome Centre as part of the Handunnetthi Group: Neuroimmune interactions. His research will focus on delineating the contribution of neuroimmune interactions to neuropsychiatric disorders, such as autism and schizophrenia. Specifically, he aims to use bioinformatic techniques and cellular modelling to investigate how viral infections can influence the way the brain develops and changes during gestation and in adulthood. Understanding how infectious agents and the brain interact will help to identify risk factors and targetable pathology in these patients, as well as inform healthcare policy relevant to their care.

Single-cell immune survey identifies a novel pathogenic role for T cells in anti-NMDA receptor encephalitis

Thomas Johnson[1]; Andrew J. Kwok[2]; Babak Soleimani[1,4]; Bo Sun[4]; Andrew Fower[4]; Mateusz Makuch[4], Julian C. Knight[1]; Ho Ko[2]; Belinda Lennox[3]; Sarosh Irani[4]; Adam E Handel[4]; Lahiru Handunnetthi[1,3,4].

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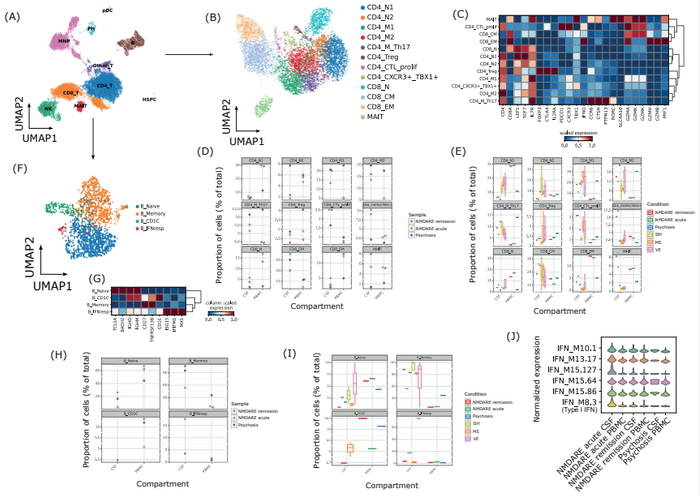
Background & Aims

- Patients presenting with N-methyl-D-aspartate receptor antibody encephalitis (NMDARE) develop psychotic symptoms, abnormal movements and seizures.
- It has been hypothesised that a subgroup of patients with only psychotic symptoms could represent a *forme fruste* of NMDARE or autoimmune psychosis (AP).
- Although autoantibodies are considered to be the key drivers of disease pathogenesis, very little is known about the contribution of immune cells other than B cells.
- In this study, we aimed to carry out a deep immune cell survey via single-cell RNA (scRNA) and adaptive immune receptor repertoire sequencing of an NMDARE patient during acute relapse and remission states, as well as an AP patient with anti-NMDAR antibodies. We aimed to contextualise our findings by leveraging publicly available scRNA data from a diverse range of neurological disease controls.

Results 2: Fine-resolution analysis of T and B cells



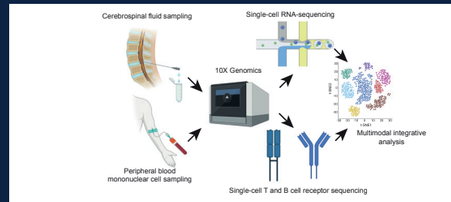
Fine-resolution analysis of T and B cells. (A) Subsetting, re-processing and reclustering of T and B cells. (B) UMAP of peripheral blood and cerebrospinal fluid T cells after reclustering. (C) Marker gene heatmap for T cell subsets. (D) Proportions of T cells in CSF and PB of NMDARE and AP. (E) Proportions of T cells from NMDARE and AP samples as compared to CSF samples from VE, MS and IIH. (F-I) Results for B cells, as described in B-E. (J) Normalised expression of six whole-blood transcriptome-derived interferon gene module in our patient samples.

Discussion

- We present a small-scale peripheral blood and CSF single-cell transcriptomic and immune repertoire atlas from patients with anti-NMDAR antibody-mediated pathology. Importantly, we found that immune profiles differed between the NMDARE and AP patients, as well as between relapse and remission states of NMDARE.
- We highlight a novel role for T cells in NMDARE pathogenesis. Specifically, we noted clonal expansion in both CD8+ and CD4+ T cells. The CD8+ effector memory cells displayed the greatest expansion with heightened cytotoxic and chemotaxic characteristics during acute relapse. Further, the antigen specificity of NMDARE T-cell clonotypes highlights a potential mechanism through which viral infections could contribute to disease mechanisms.
- Another key finding was the appearance of IFN-responsive B cells in the CSF compartment particularly during the acute phase of NMDARE.
- These results provide new insight into disease mechanisms, but larger studies are needed to confirm the generalisability of our findings.

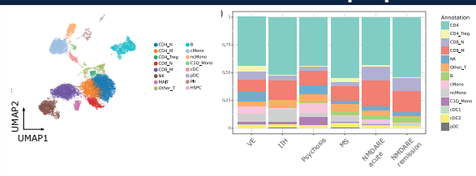
References: Kwok *et al* (2024) Preprint DOI: 10.1101/2024.09.01.24311959

Methods



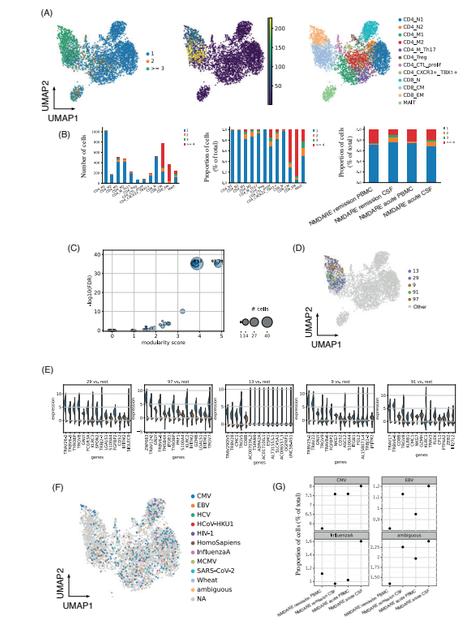
We first performed single-cell RNA and adaptive immune receptor repertoire sequencing of an NMDRE patient in relapse and remission states, as well as an AP patient with anti-NMDAR antibodies. The immune cells were loaded onto the Chromium 10X platform for library generation, and subsequently sequenced using the NovaSeq6000 platform. Next, we re-analysed publicly available cerebrospinal fluid (CSF) single-cell sequencing data from other neurological disorders: 4 viral encephalitis (VE), 3 multiple sclerosis (MS) and 5 idiopathic intracranial hypertension (IIH) .

Results 1: Cell annotations and proportions



Immune cell populations across disease states. We sequenced a total of 17,110 cells and recapitulated key mononuclear cell (MNC) types (left panel). There was concordance between our manual annotations and results from data-driven approaches. We noted several clear proportional differences in cell populations across disease states (right panel); e.g., there were more CD8+ memory (CD8_M) cells in CSF in acute compared to remission NMDARE.

Results 3: T cell receptor and clonal expansion



T cell receptor and clonal expansion analysis in NMDARE. (A) UMAPs of T cells coloured by T cell clonal expansion by clone ID size (left and middle) and T cell subset annotation (right). (B) Clonally expanded T cells in absolute numbers (left), as a total proportion of each subset (middle) and as a total proportion of each sample (right). (C) Modularity scores were calculated to test whether any of these expanded T cell clones exhibited similarities in gene expression profiles. (D) Top 5 clonotypes of highest modularity on UMAP. (E) Top differentially expressed genes between top 5 clones of highest modularity vs. other CD8 memory T cells. (F) Antigen specificity of T-cell clonotypes in NMADRE was assessed using a curated repository. (G) Proportions of viral antigens recognised by T cells in each sample.

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Clinical and laboratory predictors of poor outcomes following infectious encephalitis: systematic review and meta-analysis

Dr Thomas Johnson is a Foundation Year 1 Doctor undertaking an Academic Specialised Foundation Programme in Oxfordshire. Alongside his clinical training at Oxford University Hospitals, he is undertaking research at the Wellcome Centre as part of the Handunnetthi Group: Neuroimmune interactions. His research will focus on delineating the contribution of neuroimmune interactions to neuropsychiatric disorders, such as autism and schizophrenia. Specifically, he aims to use bioinformatic techniques and cellular modelling to investigate how viral infections can influence the way the brain develops and changes during gestation and in adulthood. Understanding how infectious agents and the brain interact will help to identify risk factors and targetable pathology in these patients, as well as inform healthcare policy relevant to their care.

Clinical and laboratory predictors of poor outcomes following infectious encephalitis: systematic review and meta-analysis

Thomas Johnson[1], Babak Soleimani[1,3], Laurissa Havins[1,3], Gregory Holt[1,2], Annapoorna Kannan[1], Isobel Howard[4], Defne Saatci[5] and Lahiru Handunnetthi[1,2,3].

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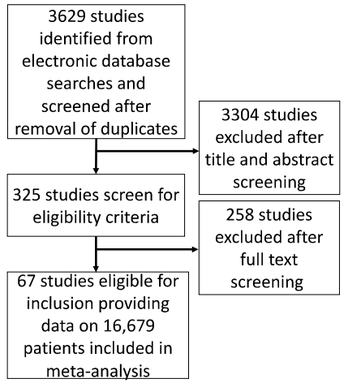
Background & Aims

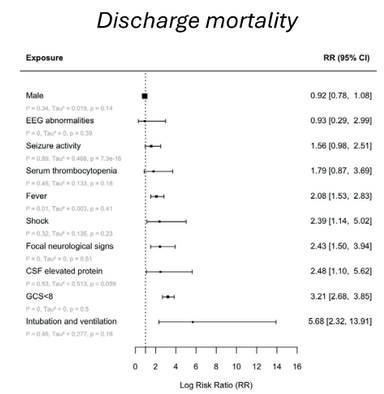
- Infectious encephalitis is a serious global health problem with high rates of mortality and subsequent health complications[1].
- Health complications following infectious encephalitis range from cognitive impairment to epilepsy[2].
- Despite this high disease burden, our ability to prognosticate mortality and morbidity following infectious encephalitis is limited.
- Accordingly, we carried out a systematic review and meta-analysis of prognostic factors following infectious encephalitis.

Methods



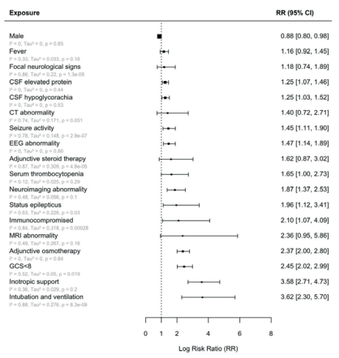
- We carried out a systematic search of MEDLINE and EMBASE from inception to 25th September 2023 (PROSPERO ID: CRD42023485045).
- Inclusion criteria were i) a primary cohort study or case series; ii) >10 participants and iv) report on clinical outcomes (mortality +/- neurological disability using using scales such as Glasgow Outcome Scale, Modified Rankin Scale or Liverpool Outcome Scale) within a 5-year period.
- We assessed i) quality using NOS and QUIPS tool, ii) heterogeneity using I^2 statistic, and iii) publication bias using funnel plot or Egger's test.
- A generic inverse variance random effects model was used to calculate pooled risk ratios for prognostic factors in Review Manager 5.4 and R package 'meta'.

Discharge mortality

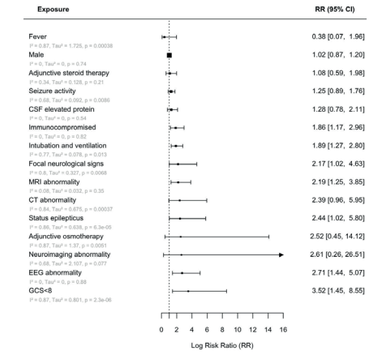


Results

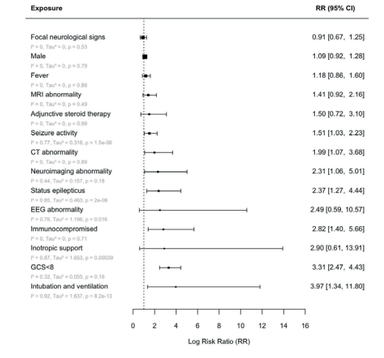
Poor outcome: discharge



Poor outcome: 1-6 months



Poor outcome: >6 months



Discussion

- This is the first comprehensive systematic review and meta-analysis of risk factors, inclusive of clinical signs, laboratory and radiological investigations, associated with mortality and neurological outcomes following infectious encephalitis.
- Focal neurological signs, elevated CSF protein and low GCS and were associated with mortality and poor outcomes at discharge.
- Immunocompromise, status epilepticus, and intubation and ventilation were linked to poor neurological outcomes at follow up.
- Adjunctive steroid did not influence long-term neurological outcomes both at discharge and at long-term follow up.
- These findings can help to stratify patients, allocate healthcare resources and implement appropriate rehabilitative strategies.

References:

1. Wang H, Zhao S, Wang S, et al. Global magnitude of encephalitis burden and its evolving pattern over the past 30 years. J Infect. 2022;84(6):777-787. doi:10.1016/j.jinf.2022.04.026
2. Kvam KA, Stahl JP, Chow FC, Soldatos A, Tattevin P, Sejvar J, Mailles A. Outcome and Sequelae of Infectious Encephalitis. J Clin Neurol. 2024 Jan;20(1):23-36. https://doi.org/10.3988/jcn.2023.0240

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Multifactorial Encephalitis in the Context of Infective Endocarditis in a 40-Year-Old Male

Dr Trisha K. Usman is born and raised in Zamboanga City. She is a graduate of the internal medicine department training program of Ciudad Medical Zamboanga, Zamboanga City, Philippines. She is a graduate of Bachelor of Science in nursing at Ateneo de Zamboanga University. She studied and graduated from Our Lady of Fatima University College of Medicine in Valenzuela, Philippines.

MULTIFACTORIAL ENCEPHALITIS IN THE CONTEXT OF INFECTIVE ENDOCARDITIS IN A 40-YEAR-OLD MALE

TRISHA K. USMAN, JERNE KAZ NIELS B. PABER, MICHAEL U. ABUTAZIL, WALID A. AMIL, SEBAR S. SALA, ROSAR JANE T. ENDAYA

PATIENT'S PROFILE

THE PATIENT IS A 40-YEAR-OLD MALE WITH A HISTORY OF CHRONIC TUBULOINTERSTITIAL NEPHRITIS WHO INITIATED HEMODIALYSIS ONE MONTH AGO, RECEIVING TREATMENT TWICE WEEKLY. HE REMAINS COMPLIANT WITH HIS DIALYSIS SESSIONS AND MEDICATIONS.

INTRODUCTION

INFECTIVE ENDOCARDITIS (IE) IS AN INFECTION OF THE HEART'S INNER LINING, OFTEN AFFECTING VALVES OR INTRACARDIAC DEVICES. RISK FACTORS INCLUDE PRIOR IE, HEART DISEASE, IV DRUG USE, IMMUNOSUPPRESSION, AND RECENT SURGERY. IE MAY PRESENT ACUTELY OR AS A CHRONIC ILLNESS WITH SYMPTOMS LIKE LOW-GRADE FEVER. DIAGNOSIS RELIES ON DUKE'S CRITERIA, INCLUDING BLOOD CULTURES, ECHOCARDIOGRAPHY, FEVER, AND RISK FACTORS. TREATMENT INVOLVES ANTIBIOTICS, VALVE FUNCTION MONITORING, REMOVAL OF INFECTED DEVICES, AND ASSESSING THE NEED FOR VALVE SURGERY.

LABORATORY RESULTS

BLOOD CHEMISTRY

- PROCALCITONIN: 94.580 NG/ML ▲
- CREATININE: 1207.8 UMOL/L ▲
- BUN: 28.1 MMOL/L ▲
- PRO BNP: 2920.0 PG/ML ▲

BLOOD CULTURE

- 06/04: (+) MRSA (HEAVY GROWTH)
  - RIGHT ARM
- 06/07: (+) MRSA (HEAVY GROWTH)
  - RIGHT LEG

2D-ECHO

- AORTIC CALCIFIC SCLEROSIS WITH VEGETATION
  MODERATE AORTIC REGURGITATION
- MITRAL VALVE SCLEROSIS WITH MODERATE MITRAL VALVE REGURGITATION

CRANIAL CT SCAN

- PLENTY SUBARACHNOID HEMORRHAGE, LEFT FRONTO-TEMPORO-PARIETAL SULCI
  MINIMAL SUBARACHNOID HEMORRHAGE, RIGHT PARIETAL SULCI
- RIGHT CEREBELLAR INFARCTION

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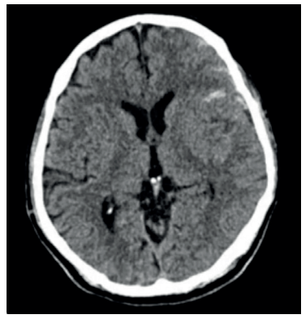
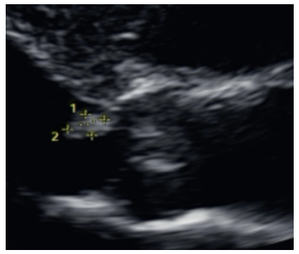
CASE DISCUSSION

THE PATIENT PRESENTED WITH A 40°C FEVER, BODY MALAISE, FRONTAL HEADACHE, AND PRODUCTIVE COUGH WITH WORSENING SHORTNESS OF BREATH OVER THREE DAYS. SUSPECTING AN INFECTION FROM HIS INTERNAL JUGULAR CATHETER, HIS PHYSICIAN RECOMMENDED ITS REMOVAL DUE TO A HISTORY OF CATHETER-RELATED BLOODSTREAM INFECTION. UPON ADMISSION, HE VOMITED, AND HIS HEADACHE WORSENED. THIS WAS FOLLOWED BY A DECREASE IN SENSORIUM AND SUDDEN VISION LOSS OF BOTH EYES.



PLAIN CRANIAL CT SCAN REVEALED SUBARACHNOID HEMORRHAGES AND LACUNAR INFARCTS. THE NEUROLOGIST SUSPECTED NON-CONVULSIVE STATUS EPILEPTICUS AS THE CAUSE OF HIS TRANSIENT BLINDNESS, ACCOMPANIED BY WORSENING SENSORIAL CHANGES.



2D-ECHO SHOWED VEGETATIONS ON THE AORTIC VALVE WITH MODERATE AORTIC REGURGITATION. REPEATED BLOOD CULTURES CONFIRMED MRSA, INDICATING LEFT-SIDED IE WITH A RISK OF SYSTEMIC EMBOLIZATION.

PATIENT INITIALLY PRESENTED WITH HIGH-RISK, NON-RESOLVING COMMUNITY-ACQUIRED PNEUMONIA. SUBSEQUENT CHEST X-RAY FINDINGS OF MIDDLE LOBE INFILTRATES LED TO A DIAGNOSIS OF PULMONARY TUBERCULOSIS. DESPITE TREATMENT, HIS RESPIRATORY DISTRESS PERSISTED, NECESSITATING NON-INVASIVE VENTILATION. HIS MULTIPLE SOURCES OF INFECTION ALSO CONTRIBUTED TO HIS PRESENTATION OF ENCEPHALITIS.

TREATMENT

TREATMENT FOR IE INCLUDED RENALLY-ADJUSTED DOSES OF MEROPENEM AND VANCOMYCIN, FOLLOWED BY LEVOFLOXACIN. FOR PULMONARY TUBERCULOSIS, A DAILY REGIMEN OF RIFAMPICIN, ETHAMBUTOL, AND ISONIAZID WAS INITIATED, ALONGSIDE FLUCONAZOLE TO MITIGATE FUNGAL RISK. HIS CARDIAC MEDICATIONS WERE CONTINUED, AND NIMODIPINE WAS GIVEN TO MANAGE NEUROLOGICAL SYMPTOMS. HEMODIALYSIS WAS MAINTAINED. RESPIRATORY SUPPORT GRADUALLY REDUCED AS HIS CONDITION IMPROVED. NOTABLY, THE ENCEPHALITIS ALSO RESOLVED WITH THE EFFECTIVE MANAGEMENT OF HIS INFECTION.

Encephalitis International — The brain inflammation non-profit

Ciudad Medical Zamboanga — MOUNT GRACE HOSPITAL

Dr Vera Fominykh

Institute of Higher Nervous Activity, Russian Academy of Sciences, (IHNA RAS) Moscow, Russia

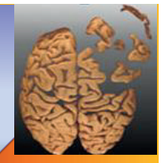
Autoimmune encephalitis in first episode of psychosis: prospective non-interventional longitudinal study in tertiary psychiatric center

From 2014 to 2022, Dr Vera Fominykh worked as a neurologist in the autoimmune department, specializing in patients with multiple sclerosis and other immune disorders at Bujanov Moscow City Clinical Hospital. Additionally, she held a researcher position at the Institute of Higher Nervous Activity and Neurophysiology during this time. In 2019, Vera successfully defended her PhD on the subject of neuroinflammation and neurodegeneration markers in autoimmune encephalitis and other immune-mediated disorders. After that, she managed many patients with AE in the country, organizing telemedical consultations and building significant expertise in treating and diagnosing AE. Together with her colleagues she worked a lot to spread the knowledge about AE, as well as to improve access to diagnostic tests. Since 2022, Vera has been working in another country, but she continues to increase awareness of AE in East-European and Central Asian Region and has recently finalized this AE study at a psychiatric institution on a volunteer basis.

Non-interventional observational study autoimmune encephalitis in first episode of acute psychosis

Vera Fominykh, Angelina Khannanova, Dmitrii Averchenkov, Lev Brylev

*work was performed at Bujanov City Clinical Hospital, Institute of Higher Nervous Activity and GBUZ «PKB N 4 DZM», Moscow, Russia
Technical and logistic work was supported by LCC "INVITRO"

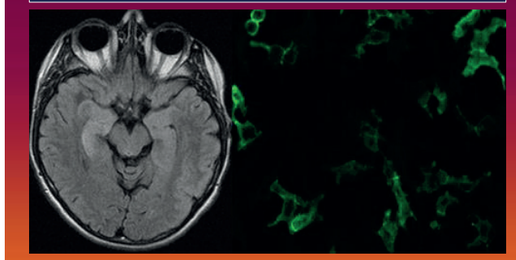


BACKGROUND AND AIMS

Autoimmune encephalitis (AE) comprises a group of non-infectious immune-mediated inflammatory disorders of the central nervous system.
The presence of antineuronal antibodies (Ab) can lead to psychiatric symptoms at the beginning of AE in 60% of patients (Herken et al., 2017).
3-7% of patients with first episode of acute psychosis (FEP) were suffered from AE (Al-Diwani et al., 2017).
These findings lead to conception of "autoimmune psychosis" as a mild form of AE (Pollak et al, 2019) as well criteria for probable, possible and definite autoimmune psychosis.
Due to the severe course of AE without treatment with high death risk and cases of post-mortem AE diagnosis in psychiatric wards, early AE diagnosis is essential.
The study aimed to reveal and characterize the AE group in FEP in a tertiary psychiatric hospital in East European settings.
Additionally, we assessed results by existing AE and autoimmune psychosis (AP) criteria.

MATERIALS AND METHODS

This study was approved by Ethics Commission (17.03.2020) and performed from June 2020 until June 2023. All FEP patients referred to the psychiatric hospital were assessed.
Inclusion criteria were age 18-60, first FEP < 1 year ago, possibility to sign the informed consent and PANSS < 5. Acute intoxication, brain trauma, lesion, infection (including COVID19 acute stage), pregnancy, other neurological pathology were exclusion criteria.
Serum and CSF were taken in the acute stage of disorders. Ogbs, antineuronal Abs to intracellular antigens, NMDA, CASPR2, LGi1, GABAb, AMPA1,2, GAD Abs, thyroid serology, ANA, dsDNA Abs were performed. CSF cytosis and protein level were tested on a regular basis. After "red flags" and analyses assessment at the first stage, the second set of analyses was performed in "positive patients": CSF antineuronal antibodies, MRI, EEG, oncological screening. If AE diagnosis was confirmed, we used immunosuppressive treatment according to the current recommendations. Outcome was assessed immediately after treatment, and after 9-12 months follow-up.
Patients screening procedure was described in the study profile.



Patient #3.
GABAb–limbic encephalitis with acute psychosis.
MRI and CSF immunoreactivity in biochips with HEK-cells (Euroimmune, Germany)

CONTACT INFORMATION

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autoimmune.neurology@gmail.com

STUDY PROFILE

784 patients admitted to Psychiatric Hospital consecutively assessed

Included: 143 patients after signing of informed consent

634 patients failed screening.
Main causes: duration of symptoms > 1 year (49 %), refuse to sign consent (28 %), acute COVID infection (11 %).
Excluded after inclusion during investigations: n=7, another diagnosis (HIV, drug use, neurosyphilis, symptoms > 1 year)

In all patients lumbar puncture was performed.

Clinical data: psychotic and neurological status, routine analysis, PANSS, cognitive assessment
«Basic» analysis:
- Serum: antineuronal Ab (hu, ma2, ri, AMPH, yo-1, CV2), NMDA, CASPR2, LGi1, GABAb, AMPA1,2, thyroid serology, ANA, antibodies to dsDNA.
- CSF: oligoclonal bands, protein level, cytosis

143 patients

Analyses assessment

Second set of analyses
- CSF: antineuronal AB (screen), NMDA, CASPR2, LGi1, GABAb, AMPA1,2,
- MRI, EEG

79 patients

5 Autoimmune Encephalitis

RESULTS:

Mean age was 32 y.o., 18-63, 75 % female.Top 3 diagnoses at admission were: F23.2, F23.1 and F20.0. The median time from admission to the first symptoms was 24 days, ranging from 0 to 361 days, and patients stayed in hospital for around 1 month. 3 % of patients were admitted to ICU and 3 patients died (2 performed suicide and 1 died due to severe AE course).
79 patients were analyzed on the second panel after "red flags" assessment or positive results on the first screening stage. In 8.4 % (n=12) of patients known antineuronal Ab were detected: NMDA (n=1), AMPH (n=1), GABAb (n=2), AMPA2 (n=1), anti-yo(n=2), GAD (n=4), MA2 (n=1). In 3 patients total CSF antineuronal antibodies were detected.

We confirmed AE diagnosis in 5 patients (3.5%):
#1: AE, AMPH+, female, "stiff-person" phenotype and psychosis. She was treated with MP pulse therapy, plasma exchange, CTX. After 1 year she returned to work.
#2: AE, AMPA2+, female, hallucinatory delusional syndrome, movement disorder, mediastinal teratoma and ovarian cystadenoma. After surgery and immunosuppressive therapy, psychotic symptoms disappeared, but movement symptoms persisted.
#3: AE, GABAb+, female, acute polymorphic psychotic syndrome. She was treated with MP pulse therapy, plasma exchange, RTX. At 1-year follow-up ovarian teratoma was detected.
#4: AE with unknown Ab, male, type 2 Ogbs, cytosis, CSF antineuronal Ab. Disease started from acute psychosis. Patient died due to severe AE cause.
#5: AE, yo+, female, acute psychosis. MP pulse therapy was successful, return to work. No oncology revealed, denied a follow-up investigation.
AE cases were assessed according AE and AP criteria.

Case	Possible AE criteria	AE with known AB	"Red flags" in psychosis	AP, possible	AP, probable	AP, defined
#1, AMPH	▼	▼	▼	▼	-	-
#2, AMPA2	▼	▼	▼	▼	-	-
#3, GABAb	-	▼	▼	-	-	-
#4, antineuronal Ab CSF	▼	AE with unknown Ab	▼	▼	▼	-
#5, yo +	-	▼	-	-	-	-

Conclusion:
- We revealed 3.5 % AE in FEP cohort in East-European tertiary psychiatric hospital.
- In 1 patient no red flags were detected. In 1 patient Ab was detected only in CSF.
- AE criteria + "red flags" assessment help to reveal 4 out 5 cases.
- 1 case was found at serum screening and confirmed by CSF analysis. Criteria of AP were too conservative to reveal AE cases. That supported the necessity of valid "red flags" panel and CSF testing in FEP cohort, as well improving of AP criteria in FEP.

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Identify, Conceptualise and Provide Care – Checklist for Persons with Acute Brain Infections.

Dr Vasundharaa S Nair (MPhil, PhD) has been working with persons with various psychiatric and neurological conditions and their families for past 6 years thereby providing individual and family interventions. She holds her research interests in the field of mental health, biological disasters like ZIKA, COVID19, Multiple Sclerosis and Acute Brain Infections. She is skilled in providing care children and parenting, women and climate change, healthy aging. She is a passionate faculty, psychotherapist and researcher.

Identify, Conceptualise and Provide Care – Checklist for Persons with Acute Brain Infections and their Caregivers.

Vasundharaa S Nair[1], Priya Treesa Thomas[2], Netravathi M[2]

[1]Jindal Institute of Behavioural Sciences, OP Jindal Global University, Sonipat and [2]National Institute of Mental Health and Neurosciences, Bengaluru

Supported by NIHR | Health Protection Research Unit in Emerging and Zoonotic Infections at University of Liverpool

Brain Infections Global

Background

Brain infections are potentially treatable conditions, that can be debilitating with serious sequelae which could result in significant brain damage and subsequent disability. WHO recognizes neurological disorders as “one of the greatest threats to public health” (WHO, 2008).

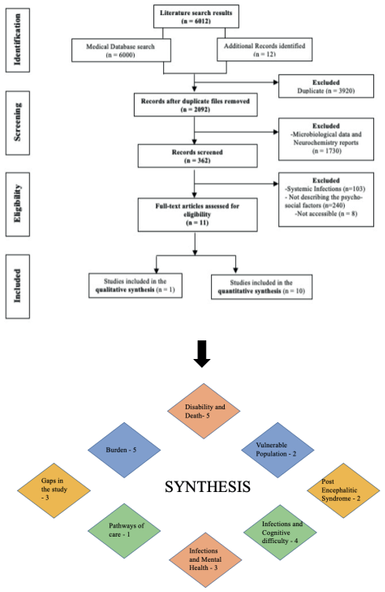


Fig 1: PRISMA-ScR and Synthesis : Areas of difficulty with respect to brain infections (Nair et.al., 2022)

Methodology

Methodology: Mixed methods study was undertaken using an exploratory sequential design.

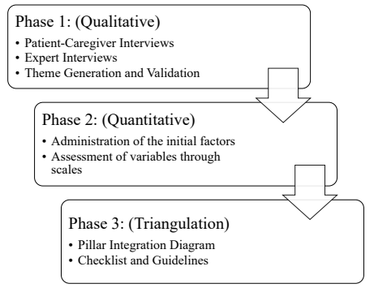


Fig 2: Research Process for the study

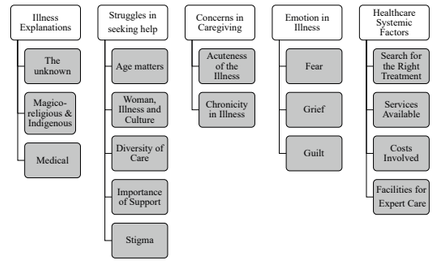
Phases

Phase 1 - Qualitative
To understand the social factors influencing care for Persons with Acute Brain Infections.

Interviews with the Patients and Caregivers	21
Interviews with Experts	15
Framework Analysis used (Ritchie & Spencer, 1994)	5 step

Phase 2 – Quantitative (n=300)
To establish the social factors which could help inform interventions for better care.

Scale Used
Semi-structured interview schedule
Initial Items of the checklist
Glasgow Coma Scale
Modified Kuppuswamy Scale
The pathway study: Encounter Form
Patient centered health services in India
Client Sociodemographic and Service Receipt Inventory



Phase 3 - Key Informant Interviews
- Triangulation
- Pillar Integration Diagram

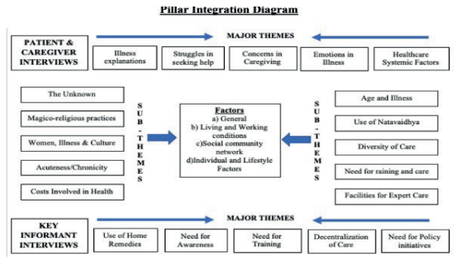


Fig 3: Triangulation

Results and Discussion

Sl. No	Domain Name	Factor	Item Description (Total 44-31)	Rating Yes – 1 No – 0
1.	Individual and Lifestyle Factors	Factor 1	My Gender makes a difference in getting help for ABI.	
			The type of job decides the care I seek for ABI.	
			My karma is the reason for ABI.	
			Home Remedies can cure ABI.	
		Factor 2	My age makes a difference in getting care for ABI.	
			Getting ABI is due to my Fate.	
			The doctors carefully listened to the concerns that I said.	
2.	Social Community Network	Factor 3	My social class makes a difference in getting treatment for ABI.	
			The agricultural harvest near me has a relation to me developing ABI.	
			My Occupation can cause ABI.	
		Factor 6	When I had problems, family members/relatives took me to a traditional healer.	
			I believe that going to the hospital will result in death.	
			I feel that doing prayers and penance can cure ABI.	
			All the hospitals I have visited have provided me with proper information.	
3.	Living and Working Conditions	Factor 5	My ability to understand the health care system helps in seeking care for ABI.	
			I went to a doctor when I developed issues.	
			My health problems (DM, HTN, etc.) are the reason for ABI.	
		Factor 8	If I am exposed to different metals/chemicals, it can cause ABI.	
			I will seek treatment for the elderly but not for me.	
			My marital status makes a difference in getting care for ABI.	
4.	General	Factor 4	Having clean Air can reduce chances of having ABI.	
			Poor nourishment can cause ABI.	
			My response to the symptoms makes a difference in getting care for ABI.	
		Factor 7	I was able to get all the medicines needed for the treatment.	
			I think there is no cure for ABI.	
			I was given answers to all the questions that I had regarding ABI.	
		Factor 9	My diet has a link to me developing ABI.	
			The hospital members were very helpful to me.	
			Stress can cause ABI.	
			I feel sad knowing about ABI.	
			I have been given information on when to come back for a follow-up for ABI.	

Fig 4: Checklist for Persons and Caregivers

Fig 5: Dahlgren-Whitehead model, 1991

Conclusion

Acute Brain Infections are one of the most important yet least understood neurological conditions. The burden of the condition is still not completely well documented.

In the growing context of Patients with Acute Brain Infections and the associated Delay causing Disability and Damage, it becomes imperative, to focus on the social, cultural, economic, environmental factors.

Further work shall focus on:

These shall thereby help in understanding of the method of better managing the condition and ensuring better quality of life helping in sensitivity and positivity towards health through a multi-disciplinary approach.

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1.World Health Organization. (2008). *Closing the gap in a generation: Health equity through action on the social determinants of health: Final report of the Commission on Social Determinants of Health (CSDH)*. Geneva: WHO.

2. Nair, V. S., Thomas, P. T., & Netravathi, M. (2022). Psychosocial factors in brain infections research in the last decade: A scoping review. *Indian Journal of Community Medicine: Official Publication of Indian Association of Preventive & Social Medicine*, *47*(4), 495–500. https://doi.org/10.4103/ijcm.ijcm_321_22 .

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Ms Walaa Zakaria

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Brain MRI Longitudinal Volumetric Characteristics Associated with Outcomes of CASPR2-Limbic Encephalitis

Ms Walaa Zakaria completed her bachelors degree in Medical Imaging Sciences with a minor in Public Health at the American University of Beirut. Currently, she is pursuing a Masters in Neurobiology at the University of Pavia in Italy. As part of her masters thesis, she is undertaking an Erasmus+ Traineeship at the French Reference Center for Paraneoplastic and Neurologic Disorders and Autoimmune Encephalitis. During this traineeship, she has developed a deep interest in quantitative imaging analysis, particularly using deep learning applications for medical image segmentation in relation to autoimmune encephalitis. She previously collaborated with the institute during her bachelor's degree, along with her supervisor at the time, to perform the volumetric analysis for the paper titled "Cerebellar Ataxia With Anti-DNER Antibodies: Outcomes and Immunologic Features." She will be graduating in October from her Masters program, and following her graduation, she is excited to continue her research journey by pursuing a PhD with the same team. This opportunity will allow her to delve deeper into the field of neuroimaging and autoimmune encephalitis, contributing to significant advancements and gaining extensive expertise in this area.

Brain MRI Longitudinal Volumetric Characteristics Associated with Outcomes of CASPR2-Limbic Encephalitis and Radiologic Presentation

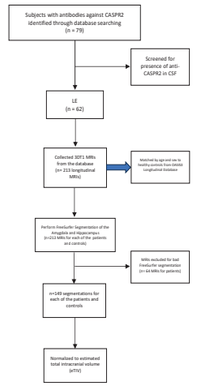
Walaa Zakaria MSc, Jeanne Benoit MD, Louis Comperat MD, Guillaume Criton MD, Antonio Farina MD, Marie Benaiteau MD, Jérôme Honnorat MD, PhD, François Cotton MD, PhD, Thomas Grenier PhD, Bastien Joubert MD, PhD

RHU BETPSY – Hospices Civils de Lyon – Université Claude Bernard Lyon, France

Introduction

Anti-CASPR2 limbic encephalitis (CASPR2-LE) is a rare autoimmune disorder mainly affecting men over 50, marked by anti-CASPR2 antibodies in cerebrospinal fluid. Symptoms, including cerebellar ataxia, seizures, and neuropathic pain, develop slowly and often correlate with MRI findings of signal changes in the medial temporal lobes and structural alterations in the hippocampus and amygdala. This study aims to investigate longitudinal volumetric changes in these brain regions associated with CASPR2-LE MRI hyperintensities to better understand their relationship with clinical outcomes, as current research on this specific condition remains limited

Methodology



The study involved a longitudinal analysis of MRI data from both CASPR2-LE patients (n=62) and healthy controls (n=62). Statistical analyses included longitudinal linear regression models to evaluate the relationship between baseline normalized hippocampus and amygdala volumes and subsequent volumes changes from baseline measured within two years and after two years from diagnosis.

Results

Normalized right and left hippocampus and amygdala volumes significantly decreased over time ($p<0.05$) (n=149 MRIs for each of the patients and controls).

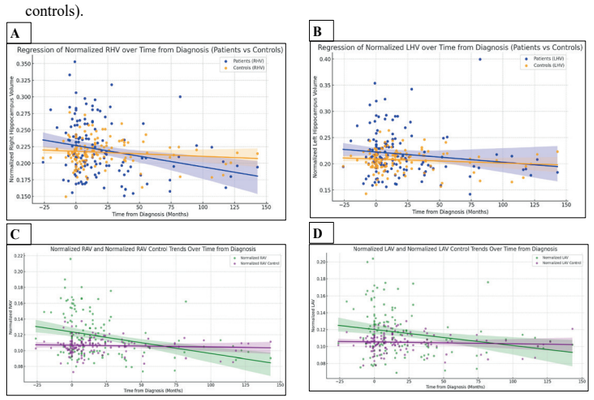


Figure 1: Time trajectory of right (A) and left (B) normalized hippocampus volumes (% eTIV) and right (C) and left (D) amygdala volumes (% eTIV) in patients and healthy controls

- **Baseline volumes** of the hippocampus (RHV, LHV) and amygdala (RAV, LAV) significantly predict volume changes over time in autoimmune encephalitis patients.
- **Within 2 years of diagnosis**, larger baseline volumes are linked to greater volume loss in both structures (hippocampus: RHV $R^2 = 0.236$, LHV $R^2 = 0.345$; amygdala: RAV $R^2 = 0.416$, LAV $R^2 = 0.465$).
- **After 2 years of diagnosis**, baseline volumes continue to predict volume changes, with stronger effects for the amygdala (hippocampus: RHV $R^2 = 0.359$, LHV $R^2 = 0.258$; amygdala: RAV $R^2 = 0.587$, LAV $R^2 = 0.653$).

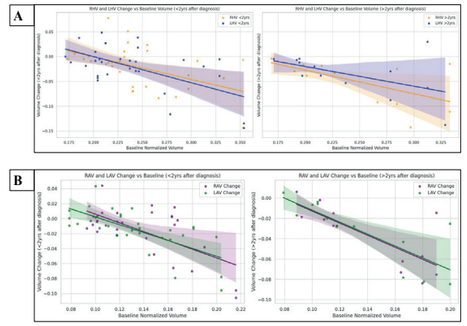


Figure 2: RHV and LHV (A) and RAV and LAV (B) Volume Changes at Followup according Baseline Volumes

Hyperintensities in the hippocampus and amygdala correlate with larger volumes, with significant differences observed at baseline in both amygdala and right hippocampus ($p<0.05$)

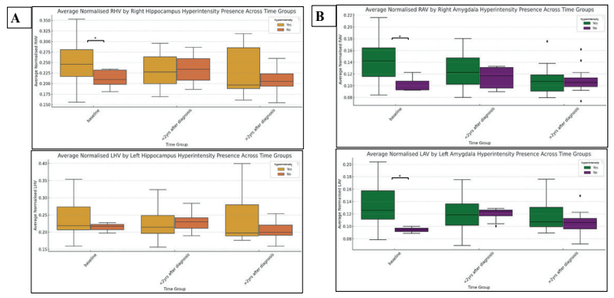


Figure 3: Comparison of Hippocampus (A) and Amygdala (B) Volumes Based on MRI Hyperintensity Status at different timepoints

Conclusions

- This study demonstrates significant longitudinal volumetric changes in the hippocampus and amygdala in patients with CASPR2 limbic encephalitis.
- Higher hippocampal and amygdala volumes at baseline are predictors of future atrophy.
- Hyperintensities in the hippocampus and amygdala correlate with larger volumes at baseline, likely reflecting active disease processes, but this relationship diminishes over time as hyperintensities may subside and be replaced by chronic atrophic changes.
- Overall, these findings enhance our understanding of the structural implications of CASPR2-LE and may inform clinical decision-making and patient management strategies.

ENCEPHALITIS CONFERENCE 2024

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Autoimmune-associated seizures disorders Modulated by IVIg – effects on Cortical Excitability: the AMICE study

Dr Yvette Crijnen works as Neurology Resident at the Neurology Department of the Erasmus University Medical Centre in the Netherlands. She combines her residency in Neurology with research in the field of autoimmune encephalitis. Her research is mainly focused on autoimmune seizures and anti-IgLON5 disease.

Effect of IVIg in autoimmune-associated seizures; the AMICE study

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Introduction

Autoimmune-associated seizures (AAS) are most frequently refractory to anti-seizure medication, but often respond to immunotherapy. Intravenous immunoglobulins (IVIg) are used off-label to treat patients with AAS.

Methodology

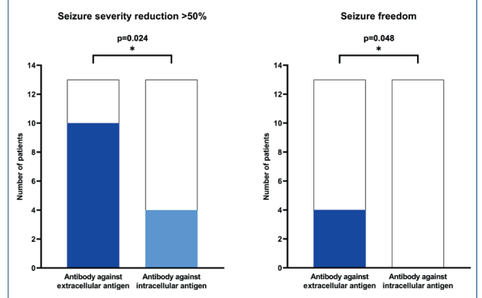
- We assessed the efficacy of IVIg in AAS
- Prospective open label clinical intervention trial with IVIg (2020-2023)
- **Treatment:** 2 courses of IVIg (Privigen): 0.4 gr/kg/day for 5 days, from day 1 and day 22
- **Inclusion criteria:** age ≥16 years, ≥1 seizure per week at baseline, antibodies in serum or CSF
- **Exclusion criteria:** alternative cause of seizures, severe encephalitis necessitating potential escalation of therapy, immunotherapy <3 months before
- **Primary outcome measures:** intention-to-treat
  - Proportion of patients with a seizure frequency reduction >50% in week 6, 7 and 8
  - Proportion of patients with seizure freedom in week 6, 7 and 8
    - For all patients, and comparison antibodies against extra- or intracellular antigen

Conclusions

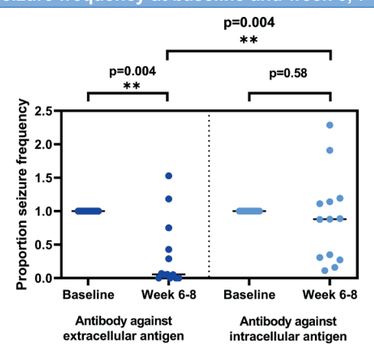
- IVIg reduces autoimmune-associated seizures
- Seizure frequency reduction, seizure severity and freedom was most seen in the patients with antibodies against an extracellular antigen
- Albeit IVIg works in AAS due to antibodies against an extracellular antigen, additional iv steroids were frequently necessary
- Delayed effects (between week 8 and 18) on seizure reduction were seen in a subset of patients with anti-GAD65 antibodies, despite lack of other additional immune therapies

Results

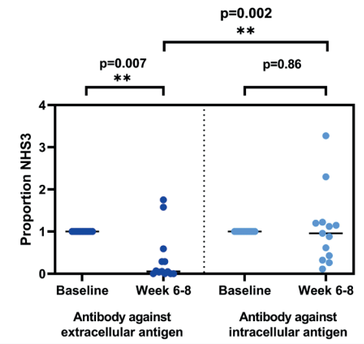
Seizure freedom and seizure reduction >50% in week 6, 7 and 8 compared to baseline



Seizure frequency at baseline and week 6, 7 and 8



Seizure severity at baseline and week 6, 7 and 8



Baseline characteristics

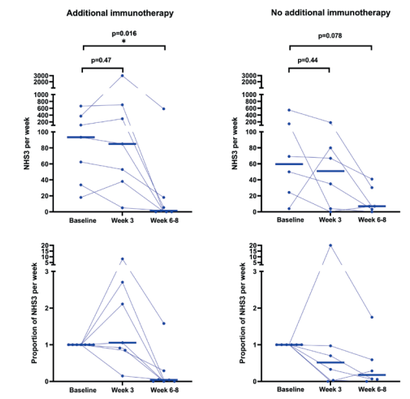
Number of patients	All patients (n=26)	Antibody against extracellular antigen (n=13)	Antibody against intracellular antigen (n=13)	*p*
Autoimmune antibody				
GAD (>10,000 IU)	13 (50%)	0 (0%)	13 (100%)	
LGI1	11 (42%)	11 (85%)	0 (0%)	
NMDAR	1 (4%)	1 (8%)	0 (0%)	
Caspr2	1 (4%)	1 (8%)	0 (0%)	
Sex, male	10 (38%)	9 (69%)	1 (8%)	0.004
Age at diagnosis, years, median (IQR, range)	48.5 (42.25-65.75, 18-75)	65 (48-71, 33-75)	43 (37-49.5, 18-62)	0.002
Seizure frequency in 3 weeks before start of IVIg, median per week (IQR, range)	18.5 (3-65.2, 1-658.3)	50.3 (18.5-232.8, 1-658.3)	3.7 (3-27.5, 2.3-73.7)	0.007
Seizure severity score (NHS3) in 3 weeks before start of IVIg, median per week (IQR, range)	31.8 (8.3-117.2, 3.3-663)	69.3 (29-256.2, 4-663)	9 (4.5-61.7, 3.3-198.7)	0.010

Follow-up characteristics

Number of patients	All patients (n=26)	Antibody against extracellular antigen (n=13)	Antibody against intracellular antigen (n=13)	*p*
Seizure freedom in week 16, 17 and 18	9 (35%)	7 (54%)	2 (15%)	0.048
Seizure severity reduction >50% in week 16, 17 and 18	22 (85%)	12 (92%)	10 (77%)	0.30
Return or increase of seizures within 18 weeks	17 (65%)	9 (69%)	8 (62%)	1.00

Additional immunotherapy <18 weeks: Antibody against extracellular antigen: n=8 steroids, n=1 steroids and rituximab, n= 1 steroids, rituximab and plasmapheresis; antibody against intracellular antigen: n=6 IVIg.

Additional vs. no additional immunotherapy before end of week 8



- 7/13 (54%) patients with LGI1 antibodies received additional steroids before the end of week 8 (median study day 22, range 14-23)
- Significant reduction of seizure severity in week 6, 7 and 8 compared to baseline in patients who received additional steroids (p=0.016)

This study was funded by the Dutch Epilepsy foundation (EpilepsieNL, 19-08) and the Interlaken Leadership Award, an unrestricted research grant from CSL Behring.

ENCEPHALITIS CONFERENCE 2024

Dr Yun Huang

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A prospective case-control study of neuroinflammation post COVID-19

Dr Yun Huang is an Academic Clinical Fellow (NIHR) in Neurology and a Specialist Registrar in Neurology, working at The Walton Centre, Liverpool. She is interested in neuroimmunology, and specifically, antibody-mediated autoimmune neurological disorders. She studied medicine at University of Cambridge (Gonville and Caius College). During her medical degree, she completed an intercalated degree in physiology, development and neuroscience. Subsequently, she completed a PhD as part of the MB PhD programme at University of Cambridge, supervised by Professor Azim Surani. Her PhD focused on exploring the molecular mechanisms underpinning key aspects of early mammalian embryonic development, the maintenance of pluripotency and primordial germ cell specification. She optimised protocols including single-cell RNA sequencing genome-wide CRISPR screening. She completed her academic foundation programme in London and moved to Liverpool as an ACF in 2021.

A prospective case-control study of neuroinflammation post COVID-19

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INTRODUCTION

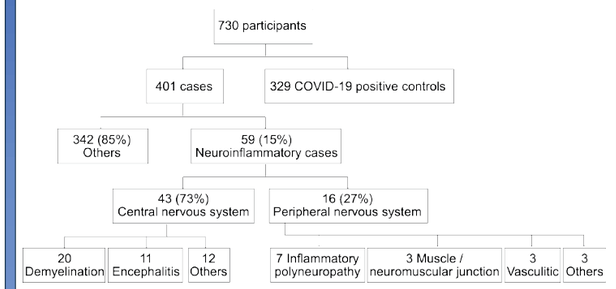
Neurological complications are common in patients post COVID-19. Central inflammatory conditions accounted for 9.3% of neurological conditions in hospitalised COVID-19 patients in a United Kingdom wide surveillance study[1]. Whilst severe neurological complications such as encephalitis are rare, they are associated with significant long-term disability and high mortality. The underlying pathophysiology remains unknown. Current knowledge is based on case studies and case series only, without a control cohort. This is the first case-control study to characterise patients who have developed neuroinflammatory complications post COVID-19.

METHODS

We carried out a prospective COVID-19 Clinical Neuroscience (COVID-CNS) study, across 17 UK sites between March 2021 and October 2022. Adults hospitalised with COVID-19, who fulfilled neurological or neuropsychiatric diagnostic criteria were recruited as cases, while controls were patients hospitalised with COVID-19 who did not develop neurological complications [2]. Participant's acute admission data, clinical follow-up assessments, blood sampling and neuroimaging were collected for analyses. Additional neuronal autoantibody assays were performed based on clinical information. Brain injury and cytokine markers were quantified using Quanterix Simoa kit and Bio-Rad human cytokine screening 48-plex kit respectively.

RESULTS

Inflammatory complications predominantly affects the central nervous system



- 59 out of 401 cases were identified as having developed a neuroinflammatory complication post COVID-19. A further 329 participants were recruited as COVID-19 positive controls
- Around 3/4 of these neuroinflammatory cases affects the central nervous system (CNS)
- CNS syndromes associated with COVID-19 are predominantly demyelinating. These includes myelitis, acute disseminated encephalomyelitis (ADEM), myelin oligodendrocyte glycoprotein antibody-associated disease (MOGAD) and clinically isolated syndrome or multiple sclerosis.
- Cases of COVID-19 associated encephalitis are mostly antibody-negative. These included 7 cases of possible autoimmune encephalitis (AE) and 4 cases of probable or definite AE as per diagnostic criteria [3]
- 4 cases of opsoclonus myoclonus syndrome were among the CNS 'Others' category
- Peripheral nervous system inflammation and vasculitis may occur following COVID-19

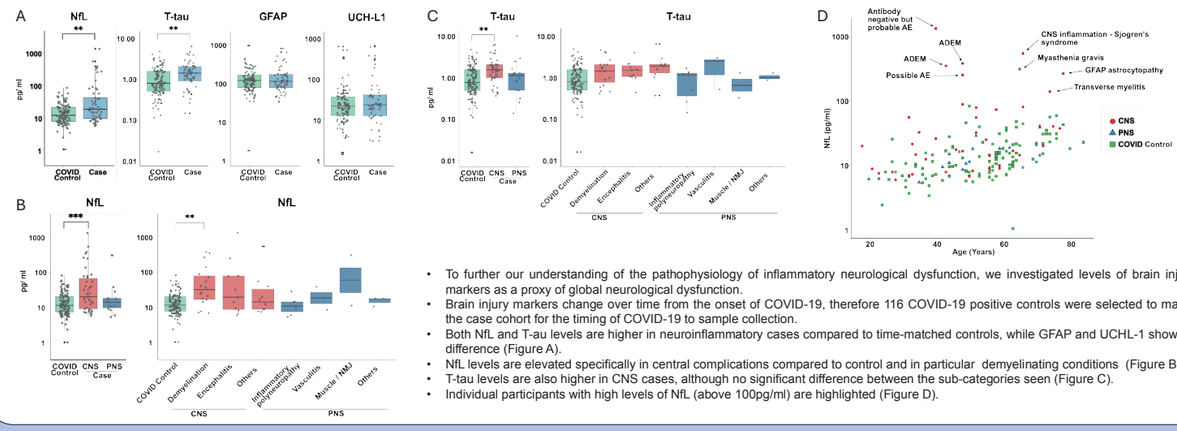
Inflammatory complications are associated with reduced Glasgow Coma Score and worsening functional outcome despite milder COVID-19

	Univariate analysis				Multivariate analysis	
Risk factor	Case (n=59)	COVID-19 positive control (n=329)	P-values	Odd's ratio	Adjusted P-value	Adjusted Odd's ratio (95% CI)	Significance
Age (median, IQR)$	50.0 (40.0 - 60.5)	57.0 (45.0 - 65.0) (NA = 1)	**0.0095**	NA			
Age group >55 (%)	64.4 (38/59)	44.2 (144/326)	**0.0042**	2.287 (1.286 - 4.068)	0.3232	1.631 (0.6179 - 4.3056)	ns
Gender (% male)	61.0 (36/59)	53.2 (175/329)	0.2765	1.358 (0.777 - 2.411)			
Clinical frailty score (median, IQR)$	2.0 (1.5 - 2.0) (NA = 4)	2.0 (2.0 - 3.0) (NA = 79)	0.2636	NA			
Ethnicity (% white)	84.3 (43/51, NA = 8)	78.4 (225/287, NA = 42)	0.3367	0.675 (0.302 - 1.511)			
Co-morbidities							
Hypertension (%)	23.2 (13/56, NA = 3)	33.3 (104/329, NA = 17)	0.1343	0.605 (0.312 - 1.174)			
Tobacco smoking (%)	10.6 (5/47, NA = 12)	5.1 (15/295, NA = 34)	0.1318	2.222 (0.768 - 6.432)			
Hypercholesterolaemia (%)	3.7 (2/54, NA = 5)	14.5 (45/311, NA = 18)	**0.0292**	0.227 (0.053 - 0.966)			
Diabetes (%)	17.9 (10/56, NA = 3)	17.9 (56/313, NA = 16)	0.9951	0.998 (0.475 - 2.096)			
Neurological condition (%)	10.9 (6/55, NA = 4)	3.8 (12/316, NA = 13)	**0.0235**	3.102 (1.112 - 8.649)			
Psychiatric disorder (%)	10.7 (6/56, NA = 3)	8.6 (27/314, NA = 15)	0.5117	1.275 (0.501 - 3.247)			
Chronic cardiac disease (%)	1.8 (1/56, NA = 3)	5.7 (18/315, NA = 14)	0.2191	0.305 (0.039 - 2.294)			
Chronic kidney disease (%)	5.6 (2/56, NA = 3)	6.7 (21/314, NA = 15)	0.3736	0.517 (0.118 - 2.267)			
Chronic pulmonary disease (%)	14.5 (8/55, NA = 4)	14.3 (45/315, NA = 14)	0.9595	1.021 (0.453 - 2.303)			
Autoimmune disease (%)	1.9 (1/54, NA = 5)	4.1 (13/315, NA = 14)	0.4188	0.438 (0.056 - 3.421)			
Pre-admission medications							
Immunomodulatory medication (%)	9.4 (5/53, NA = 6)	3.9 (12/307, NA = 22)	0.0799	2.561 (0.854 - 7.593)			
Steroid (%)	16.0 (8/50, NA = 9)	20.5 (63/307, NA = 22)	0.4576	0.738 (0.330 - 1.650)			
Anti-epileptic medication (%)	9.6 (5/52, NA = 7)	2.6 (8/306, NA = 23)	**0.0126**	3.963 (1.244 - 12.628)			
Ant-depressants (%)	11.8 (6/59, NA = 8)	10.2 (31/305, NA = 24)	0.7288	1.178 (0.465 - 2.985)			
Anti-platelets (%)	4.2 (2/48, NA = 11)	7.9 (23/292, NA = 37)	0.3614	0.509 (0.119 - 2.230)			
Anti-coagulants (%)	5.9 (3/51, (NA = 8)	7.4 (23/309, NA = 20)	0.6899	0.777 (0.225 - 2.689)			
ACE inhibitor (%)	17.3 (9/52, NA = 7)	11.5 (35/305, NA = 24)	0.237	1.615 (0.725 - 3.594)			
Statin (%)	11.5 (6/52, NA = 7)	20.7 (63/305, NA = 24)	0.1238	0.501 (0.205 - 1.226)			
Admission characteristics							
GCS on admission (mean, SD)$	13.5, 3.1 (NA = 20)	14.9, 0.6 (NA = 61)	**<0.0001**	NA			
GCS on admission <15 (%)	25.6 (10/39, NA =20)	1.9 (5/268, NA = 61)	**<0.0001**	18.138 (5.801 - 56.711)	**0.0082**	**8.2539 (1.7277 - 39.4316)** **	
Respiratory symptoms (%)	54.2 (32/59)	84.8 (279/329)	**<0.0001**	0.212 (0.117 - 0.385)			
Fever (%)	32.2 (19/59)	55.3 (182/329)	**0.0011**	0.384 (0.213 - 0.691)			
COVID severity score on admission (mean, SD)$	3.1, 2.7 (NA = 6)	4.5, 1.4 (NA = 4)	**<0.0001**	NA			
COVID severity score on admission (1-3) (%)	54.7 (29/53, NA = 6)	16.3 (53/325, NA = 4)	**<0.0001**	0.161 (0.0871 - 0.299)	**5.26x10^{-05}**	**0.1432 (0.05585 - 0.3674)** ***	
Worst COVID severity score during admission (mean, SD)$	3.8,2.5 (NA = 16)	4.8, 1.5 (NA = 13)	**0.0002**	NA			
Outcome							
Activities of daily living (median, IQR) $	3.0(0.0-4.1)(NA=19)	1.0 (0.0-2.8) (NA = 135)	**0.0015**	NA			
Estimated mRS >1 (%)	72.5 (29/40, NA = 19)	52.1 (151/194, NA = 135)	**0.0179**	2.439 (1.148 - 5.134)	**0.0248**	**3.2572 (1.1616 - 9.1338)** *	

$ Mann-Whitney two-sided, all other statistical analyses were Chi-squared two-sided test. NA = data not available.

- We used a multivariate analysis model to compare a range of risk factors between neuroinflammatory cases and COVID-19 positive controls.
- We found a Glasgow Coma Score (GCS) of less than 15 on admission and an estimated modified Rankin Score >1 were associated with a higher risk of developing inflammatory neurological complications, while a COVID severity score of between 1-3 on admission was associated with a lower risk.

Axonal and dendritic injury markers are elevated in central inflammatory conditions



- To further our understanding of the pathophysiology of inflammatory neurological dysfunction, we investigated levels of brain injury markers as a proxy of global neurological dysfunction.
- Brain injury markers change over time from the onset of COVID-19, therefore 116 COVID-19 positive controls were selected to match the case cohort for the timing of COVID-19 to sample collection.
- Both NfL and T-tau levels are higher in neuroinflammatory cases compared to time-matched controls, while GFAP and UCHL-1 show no difference (Figure A).
- NfL levels are elevated specifically in central complications compared to control and in particular demyelinating conditions (Figure B)
- T-tau levels are also higher in CNS cases, although no significant difference between the sub-categories seen (Figure C).
- Individual participants with high levels of NfL (above 100pg/ml) are highlighted (Figure D).

CONCLUSIONS

- Neuroinflammatory complications post COVID-19 predominately affects the central nervous system
- Neuroinflammatory cases are associated with reduced GCS and poor functional outcome despite having a milder COVID-19 on admission
- We found elevated levels of NfL and T-tau in central neuroinflammatory complications
- We aim to further investigate the role neuronal auto-antibodies plays in the pathogenesis of neuroinflammatory complications post COVID-19

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ACKNOWLEDGEMENTS

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THE COVID-CLINICAL NEUROSCIENCE STUDY
UNIVERSITY OF LIVERPOOL
NIHR | National Institute for Health and Care Research
Yun Huang is an NIHR academic clinical fellow in Neurology

Encephalitis International Scientific Advisory Panel

Encephalitis International
The brain inflammation non-profit