



Guilty by Association: KCTD16 and GABA_BR Antibodies in Paraneoplastic Limbic Encephalitis

The Expanded Clinical Spectrum of Anti-GABA_BR Encephalitis and Added Value of KCTD16 Autoantibodies

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In this study, we report the clinical features of 32 patients with γ -aminobutyric acid B receptor (GABA_BR) antibodies, identify additional autoantibodies in patients with anti-GABA_BR encephalitis that mark the presence of an underlying small-cell lung carcinoma and optimize laboratory methods for the detection of GABA_BR antibodies. Patients (n = 3225) were tested for the presence of GABA_BR antibodies using cell-based assay, immunohistochemistry, and live hippocampal neurons. Clinical data were obtained retrospectively. Potassium channel tetramerization domain-containing (KCTD)16 antibodies were identified by immunoprecipitation, mass spectrometry analysis and cell-based assays. Potassium channel tetramerization domain containing 16 antibodies were identified in 23 of 32 patients with anti-GABA_BR encephalitis, and in 1 of 26 patients with small-cell lung carcinoma and Hu antibodies, but not in 329 healthy patients and disease controls. Of the anti-GABA_BR encephalitis patients who were screened sufficiently, 18 (95%) of 19 patients with KCTD16 antibodies had a tumor versus 3 (33%) of 9 anti-GABA_BR encephalitis patients without KCTD16 antibodies ($P = .001$). In most cases, this was a small-cell lung carcinoma. Patients had cognitive or behavioral changes (97%) and prominent seizures (90%). Thirteen (42%) patients developed a refractory status epilepticus with intensive care unit admittance. Strikingly, 4 of 32 patients had a rapidly progressive dementia. The addition of KCTD16 to the GABA_BR cell-based assay improved sensitivity of the in-house fixed cell-based assay, without loss of specificity. Twenty-two of 26 patients improved (partially) to immunotherapy or chemotherapy. Anti-GABA_BR encephalitis is a limbic encephalitis with prominent, severe seizures, but patients can also present with rapidly progressive dementia. The co-occurrence of KCTD16 antibodies points toward a paraneoplastic origin. The addition of KCTD16 improves the sensitivity of the cell-based assay.

Commentary

Since its seminal description in the 1960s,¹ the spectrum of limbic encephalitis (LE) has considerably expanded. So has our understanding of its causes, with the establishment of a link with cancer and the identification of onconenural antibodies in the 1990s, followed by the identification of sporadic autoimmune cases and of antibodies to neuronal surface antigens in the 2000s. There are currently more than a dozen antibodies associated with idiopathic and paraneoplastic LE (Table 1). The γ -aminobutyric acid B receptor (GABA_BR) antibody is a relatively recent addition to this expanding list. With around 150 cases reported prior to this series²⁻¹¹ (Table 2), this entity is rare—approximately 20 times less frequent than the anti-NMDAR syndrome,² for instance—although it is likely underdiagnosed.

In this study, the authors report the clinical features of a new series of 32 patients diagnosed at a national reference center. They also describe a novel antibody targeted at the potassium

channel tetramerization domain-containing protein 16 (KCTD16) in a subgroup of these patients, especially paraneoplastic cases.

Most (27/32) of these new patients with a GABA_BR antibody presented with LE, and seizures occurred in all. Seizures were the first clinical manifestation in 17 patients and evolved to refractory status epilepticus in 13. The most frequent seizure semiology was generalized or bilateralized tonic-clonic, although some patients also had focal seizures with impairment of consciousness, with brain magnetic resonance imaging and electroencephalography pointing toward unilateral or bilateral mesial temporal lobe foci. Four patients presented with rapidly progressive dementia, which mimicked the clinical and biological presentation of Creutzfeldt-Jakob disease, including the elevation of 14-3-3 and τ protein levels in the cerebrospinal fluid (CSF). Whether or not subclinical seizures might have contributed to the rapid cognitive decline was not systematically investigated. One patient had subacute cerebellar ataxia.



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Table 1. List of Antibodies Linked With Limbic Encephalitis, With Their Specific Clinical Features and Association With Neoplasms.

Antibody	Clinical Features	% Paraneoplastic (Associated Neoplasms)
Intranuclear and cytoplasmic antigens		
Hu	Rarely isolated LE; often signs of widespread nervous system involvement	>90% (lung [mostly SCLC], breast, melanoma, prostate)
Ma2/Ta	Isolated LE or LE combined with cerebellar degeneration or brainstem encephalitis	>90% (testicular)
CV2/CRMP5	Isolated LE, or in association with signs of widespread nervous system involvement.	>90% (lung, thymoma)
Intracellular synaptic antigens		
Amphiphysin	Rarely isolated LE; often signs of widespread nervous system involvement	>90% (breast)
GAD65	LE, isolated epilepsy; type I diabetes	Rare (lung, pancreas, thymic carcinoma)
Neuronal surface antigens		
LGII	LE often preceded by faciobrachial tonic seizures; frequent SIADH	Rare (thymoma, lung [SCLC])
Caspr2	LE; more commonly neuromyotonia or Morvan syndrome	Uncommon (thymoma)
Adenylate kinase 5	LE	Limited data available
AMPA receptor	LE	70% (lung, breast, thymic carcinoma)
GABA _B receptor	LE with prominent seizures and SE; RPD; OMS; PERMS; myelopathy; ataxia	60% (lung [SCLC], thymic carcinoma, thymoma, bladder, breast)
Glycine receptor	LE with prominent; isolated epilepsy	Rare (lung [SCLC])
GABA _A receptor	Multifocal encephalitis with limbic, neocortical, and subcortical involvement, often with refractory SE	Uncommon (lung [SCLC], thymoma)
P/Q-type VGCC	Rarely isolated LE, often signs of widespread nervous system involvement; Lambert-Eaton syndrome	Frequent (lung [SCLC])
N-type VGCC	LE; ataxia	Frequent (lung [SCLC])
mGluR5	Isolated LE (Ophelia syndrome)	>90% (Hodgkin lymphoma)

Abbreviations: AMPA, alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid; Caspr2, contactin-associated protein-like 2; CRMP-5, collapsin response mediator protein-5; GABA, γ -aminobutyric acid; GAD65, glutamic acid decarboxylase 65 kDa; LE, limbic encephalitis; LGII, leucine-rich, glioma inactivated 1; mGluR5, glutamate metabotropic receptor 5; SIADH, syndrome of inappropriate antidiuretic hormone secretion; SE, status epilepticus; SCLC, small-cell lung carcinoma; VGCC, voltage-gated calcium channel.

This report thus further confirms that LE with prominent ictal manifestations is the most typical clinical presentation of GABA_BR antibodies. It also underscores that the range of uncommon clinical manifestations associated with the antibody extends beyond LE. In addition to rapidly progressive dementia and subacute cerebellar ataxia, cases of opsoclonus–myoclonus syndrome, progressive encephalopathy with rigidity and myoclonus, and brainstem encephalitis were previously reported.^{3–5} Importantly, the response to immune and oncological therapies was observed in 22 of 26 treated patients, with seizure control being achieved faster than cognitive improvement. Eighteen patients achieved a good outcome with a score ≤ 2 on the modified Rankin scale (mRS), although no patient became asymptomatic (mRS of 0). This indicates that most patients had residual cognitive dysfunction, at least during the duration of the follow-up period.

Systematic cancer screening with whole-body fluorodeoxyglucose-positron emission tomography (FDG-PET) led to the identification of an underlying cancer in 21 cases, most frequently small-cell lung carcinoma (SCLC), confirming prior reports of a paraneoplastic etiology in 60% of cases (Table 2). This obviously indicates that thorough screening for an occult neoplasm is mandatory these cases.

Current expert guidelines for SCLC screening recommend performing chest computed tomography (CT), followed by

FDG-PET if the CT is negative, or integrated FDG-PET/CT every 6 months for 4 years.¹² Although intensive repeated screening is certainly warranted for cases with a high risk of occult neoplasm, such as those with antibodies targeted at intranuclear or cytoplasmic antigens (Hu, Ma2/Ta, etc), it might not be cost-effective in patients with a lower risk, such as most of those with neuronal surface antibodies (LGII, Caspr2, etc). In fact, inappropriate screening might even become a source of unnecessary exposure to radiation and produce anxiety in these patients. In these cases, additional information that can help assess more accurately the probability of a paraneoplastic etiology and the need for intensive screening would be welcome. And this is where the major novelty of this study lies. The authors identified a new antibody directed at KCTD16. This protein belongs to a family of intracellular proteins that bind to the GABA_BR and contribute to the downstream inhibitory signaling of the receptor. A KCTD16 antibody was found in 23 of 32 patients with GABA_BR antibody. Most importantly, its presence was associated with a much higher risk of an underlying neoplasm (17/23 vs 3/9). The authors also identified onconeural antibodies in 2 of the KCTD16-negative paraneoplastic cases (one Hu and one voltage-gated calcium channel [VGCC]) and in 6 of the KCTD16-positive paraneoplastic cases (including Hu, SOX1, Ri, and VGCC). In previous series, other antibodies (CV2/CRMP-5, amphiphysin) had been shown to

Table 2. Summary of Clinical and Paraclinical Findings in Cases of GABA_BR Antibody-Associated Limbic Encephalitis Reported in Large (N > 5) Case Series.

Number of cases	179
Gender (male; N [%])	118 (67%)
Age (years) median (range)	62 (4-85)
Seizures as presenting symptoms (N [%])	127/167 (76%)
Seizure semiology	Generalized tonic-clonic, focal with impairment of consciousness (temporal)
Status epilepticus (N [%])	46/150 (30%)
Abnormal MRI (N [%])	84/156 (54%)
Abnormal CSF (N [%])	96/147 (65%)
Associated tumors (N)	SCLC (89), Thymic carcinoma (2), Lung, neuroendocrine (1), Thymoma (1), Breast (1), Bladder (1), Unknown type (5)
Associated autoimmune disorder (N)	Diabetes (2), Thyroid disorder (2)
Associated antibodies (N)	Hu (8), SOX1 (10), CV2/CRMP-5 (1), ANNA-3 (1), Amphiphysin (1), N-type VGCC (14), P/Q-type VGCC (4), NMDA receptor (1), VGKC complex (1), AMPA receptor (1), GABA _A receptor (1), GAD65 (9), Thyroid (3)
Response to immune therapy (N [%])	99/124 (80%)


Abbreviations: AMPA, alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid; ANNA-3, anti-neuronal nucleus antigen-3; CRMP-5, collapsin response mediator protein-5; CSF, cerebrospinal fluid; GABA, gamma-aminobutyric acid; GAD65, glutamic acid decarboxylase 65 kDa; MRI, magnetic resonance imaging; NMDA, N-methyl-D-aspartate; SCLC, small-cell lung carcinoma; VGKC, voltage-gated potassium channel; VGCC, voltage-gated calcium channel.

co-occur in a few patients with GABA_BR antibodies.²⁻⁶ Thus, the presence or absence of additional antibodies, such as KCa1.1, Hu, SOX1, or VGCC, in addition to GABA_BR antibody, has important clinical implications, as it does modify the likelihood of an underlying cancer and might help guide clinical management. As a side note, the presence of a tumor and of KCa1.1 antibody was associated with higher GABA_BR antibody titers in the CSF and a more severe clinical course. This association between the presence of an underlying tumor, a higher antibody titer and a more severe clinical picture, is reminiscent of the anti-NMDAR syndrome.

The authors also provide important information regarding the sensitivity and specificity of different methods used for antibody detection and identification. Immunostaining of

sections of cerebral issue and of live neurons had a sensitivity close to 100% on both CSF and serum. These methods however do not allow the identification of specific antibodies and are used for screening. Specific detection of GABA_BR antibodies using both commercially available and in house cell-based assays showed a similarly suboptimal (84%-97%) sensitivity. For the practicing clinician, the key point to remember is that commercially available methods lack sensitivity for the detection of known antibodies, including GABA_BR, and are unable to identify antibodies that are yet to be discovered. A negative result does not exclude the diagnosis of autoimmune encephalitis and should prompt further investigations, with the help of a research lab, if possible. Perfect sensitivity was indeed achieved using an in-house tailored cell-based assay combining multiple antigens (GABA_BR and KCTD16).

Altogether, this study further underscores the clinical and immunological complexity of LE. It also provides exciting new leads on how to better tease out idiopathic and paraneoplastic cases, and ultimately improve patient care. Hopefully, this observation will be replicated in larger studies and similar findings will emerge in cases of LE associated with other antibodies that portend a comparable or even lower overall risk of paraneoplastic etiology, such as Caspr2 or LGI1.

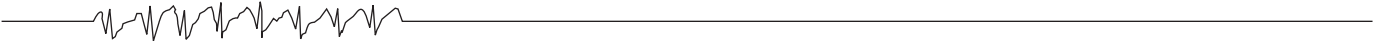
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