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P/Q- & N-TYPE CALCIUM CHANNEL ANTIBODIES: ONCOLOGICAL, NEUROLOGICAL & SEROLOGICAL ACCOMPANIMENTS

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Abstract

Introduction—Voltage-gated calcium channel-autoimmunity (VGCC-P/Q-type and VGCC-N-type) occurs beyond Lambert-Eaton syndrome and lung cancer.

Methods—We reviewed records for 236 Mayo Clinic patients with VGCC antibodies found in evaluation for paraneoplastic neurological autoimmunity (generally without myasthenic syndromes).

Results—VGCC autoantibodies were detected in 3.4% of neurological patients, 1.7% of healthy controls, and 4% of neurologically-asymptomatic lung cancer controls. Fifty neurological patients (21%) had ≥1 neoplasm, historically (46) or detected prospectively [small-cell lung carcinoma (2), breast adenocarcinoma (2), lymphoma (1), and suspected tonsillar carcinoma (1)]. Autoimmune neurological diagnosis frequencies (encephalopathy, ataxia, myelopathy, neuropathy, neuromuscular junction disorder, and myopathy) among patients with medium values (24%; 0.10–0.99 nmol/L) or low values (19%; 0.03–0.10 nmol/L) were fewer than among patients with antibody values exceeding 1.00 nmol/L (71%; P=0.02 and 0.004, respectively).

Discussion—Among neuronal VGCC-autoantibody-seropositive patients, autoimmune neurological phenotypes and cancer-types are diverse. Cautious interpretation of results (particularly medium and low values) is advised.

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Disclosures

NZ, DHL, CK reports no disclosures

VL is named inventor on a patent relating to AQP4 as NMO antigen, and a pending patent related to AQP4 and cancer. Earnings to date from licensing this technology have exceeded the federal threshold for significant interest.

SP has received no royalties to date but may accrue revenue for patents relating to AQP4 antibodies for diagnosis of neuromyelitis optica and AQP4 autoantibody as a cancer marker. He receives research support from the Guthy-Jackson Charitable Foundation, Alexion Pharmaceuticals, Inc. and the National Institutes of Health (RO1 NS065829).

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Keywords

Calcium channel; paraneoplastic; myasthenia; ataxia; neuropathy

INTRODUCTION

Voltage-gated calcium channels of P/Q-type (VGCC-P/Q) and N-type (VGCC-N) are targets of IgG-mediated motor nerve terminal autoimmunity in Lambert-Eaton syndrome (LES). Early supportive evidence included structural alterations of calcium channel-enriched presynaptic membranes in freeze-fracture electron microscopic images and disruption by LES patient serum of depolarization-dependent calcium influx in cultured small-cell carcinoma cells and murine motor nerve terminals.[1–3] The snail-derived omega neurotoxins MVIIC and GVIA specifically antagonize P/Q- and N-type calcium channels, respectively.[4,5] Radiolabeling of these high affinity peptide antagonists permitted development of radioimmunoassays for antibody detection.[4,6,7] VGCC-P/Q antibodies are detectable in more than 90% of non-immunosuppressed patients with LES, and VGCC-N antibodies are detected in 73% of paraneoplastic cases (and 36% of non-paraneoplastic cases).[6] A spectrum of VGCC-antibody-associated neurological disorders beyond LES has been supported by the Mayo Clinic Neuroimmunology Laboratory's subsequent service serological experience of patients referred for paraneoplastic evaluation.[6] Here we describe the frequency of VGCC-P/Q and VGCC-N antibody seropositivity in patients referred for paraneoplastic antibody evaluation on a service basis at the Mayo Clinic (not restricted to a neurological phenotype), as well as the frequency and spectrum of oncological, neurological, and serological accompaniments during a 24-month period. We also determined the frequency of neuronal VGCC-antibodies in serum of 2 control groups: healthy persons, and neurologically-asymptomatic patients with lung cancer.

PATIENTS & METHODS

The study was approved by the institutional review board of Mayo Clinic, Rochester, MN (IRB 14-5646). From July 1 2011 to June 30 2013, service paraneoplastic autoantibody evaluation of serum from 6842 patients seen at Mayo Clinic (Rochester Minnesota) yielded 236 positive results for VGCC-P/Q or VGCC-N antibody (3.4%).

Control subjects

Serum samples were available for: 1) 173 healthy residents of Olmsted County, Minnesota, who were of similar age and gender distribution to our patient referrals (collected in 2005), 97 were women (56%), and median age of serum draw was 52 years (range, 18–83 years); 2) 245 neurologically-asymptomatic patients with primary lung cancer [treatment-naïve, identified from the Mayo Clinic Epidemiology and Genetics of Lung Cancer Research registry, enrolled 1999 to 2005 (small-cell lung carcinoma, 35; adenocarcinoma, 70; squamous carcinoma, 70; other lung cancer types, 70)][8]; 3) 202 patients consecutively referred for the paraneoplastic evaluation, but were seronegative (2004–2005).[9]

Clinical evaluation

N.Z. and A.M. reviewed the electronic medical record of all seropositive patients. This included history and examination findings, laboratory values, imaging findings, results of electrophysiological tests (electromyography, electroencephalography, and autonomic studies) and cerebrospinal (CSF) results. N.Z. and A.M. also confirmed the neurological diagnosis made by an evaluating Mayo Clinic neurologist.

Serological evaluation

VGCC antigens were solubilized in non-denaturing detergent (digitonin) from cerebral cortical membranes (porcine for VGCC-N; combined porcine and rabbit for VGCC-P/Q) and tagged by complexing with ^{125}I–labeled ω-conopeptide GVIA or MVIIC, respectively. Antibodies were detected by radioimmunoprecipitation using goat anti-human immunoglobulin (IgG/IgM) as precipitant. Positive sera were re-evaluated in the course of this study comparing anti-human IgG/IgM and anti-human IgG as precipitants; values obtained were not significantly different from those originally obtained using a goat anti-human IgG/IgM precipitant, which are reported here. Results are expressed in terms of precipitated ^{125}I-ω-conopeptide (nanomoles per liter of serum), after subtracting the value precipitated by healthy control sera, and repeating the assay with ^{125}I-ω-conopeptide alone to eliminate false positive results.[10]

For antibody-positive patients, clinical/laboratory correlations were analyzed according to antibody value: high (VGCC-P/Q and VGCC-N, 1.00 nmol/L or greater), medium (VGCC-P/Q and VGCC-N, 0.10–0.99 nmol/L), and low (VGCC-P/Q, 0.03–0.09 nmol/L; VGCC-N, 0.04–0.09 nmol/L).[8] Values of 0.00–0.02 nmol/L (VGCC-P/Q antibody) and 0.00–0.03 nmol/L (VGCC-N antibody) are considered normal. We compared the frequency of neurological diagnoses and cancers among patients and seronegative controls, and autoimmune diagnoses among antibody value groups using the Fisher exact test ($P < 0.05$ was considered significant).

All sera were evaluated additionally at time of clinical evaluation by: 1) standardized immunofluorescence assays for IgG autoantibodies of the following specificities: anti-neuronal nuclear (ANNA)-1, -2, -3; amphiphysin; Purkinje cell cytoplasmic -1, -2, and -Tr; collapsin response-mediator protein (CRMP)-5; anti-glial/neuronal nuclear (AGNA)-1; n-methyl-D-aspartate (NMDA) receptor (GluN1 subunit); a-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid [AMPA receptor (GluA1 and GluA2)]; and gamma-aminobutyric acid (GABAB) receptor; 2) radioimmunoprecipitation assays for antibodies targeting neuronal voltage-gated potassium channel (VGKC)-complexes, muscle (α1) nicotinic acetylcholine receptor, and neuronal ganglionic (α3) acetylcholine receptor; and 3) enzyme-linked immunosorbent assay for striational antibodies. For the purpose of this study, we additionally performed cell-based assays using human embryonic kidney-293 cells transfected with appropriate expression plasmids to detect antibodies targeting NMDA receptor, AMPA receptor, and GABAB receptor (Euroimmun, Lübeck, Germany). Assays for muscle acetylcholine receptor modulating antibody, muscle-specific kinase (MuSK) antibody, leucine-rich-glioma-inactivated-1 (Lgi1) and contactin-associated protein-2

(CASPR2) IgGs, and glutamic acid decarboxylase 65-isoform (GAD65) antibody were not performed on all sera, but their detection frequencies are reported for patients tested.

RESULTS

Control subjects

Calcium channel antibodies were detected in 3 of 173 healthy control subjects (1.7%) and 10 of 245 neurologically asymptomatic patients with lung cancer (4%). Two healthy controls had VGCC-P/Q antibody (0.10 and 0.18 nmol/L), and 1 had VGCC-N antibody (0.05 nmol/L). Among the 10 lung cancer patients, 3 had VGCC-P/Q antibody (median value, 0.03 nmol/L; range, 0.03–0.52) and 8 had VGCC-N antibody (median, 0.05 nmol/L, range 0.04–0.13); 1 patient had both antibodies. Their carcinomas were squamous cell (4), non-small-cell, not otherwise specified (3), small-cell (2), and adenocarcinoma (1).

Neurological patients

One or both VGCC antibodies were detected in 236 of the 6842 patients tested (3.4%; VGCC-P/Q only, 142; VGCC-N only, 73; both, 21). None of neurological phenotype, autoimmune diagnosis, or cancer type could be predicted by calcium channel antibody profile (VGCC-P/Q, VGCC–N, or both). Fifty-two percent of the seropositive patients were women. Oncological, neurological, and serological accompaniments of seropositivity are summarized in Supplementary Table S1 (available online), and Tables 1 and 2, respectively. The median age at neurological symptom onset was 57 years (range 9–87 years). The median Mayo Clinic follow up period after initial evaluation was 7 months (range, 0 to 156 months).

Oncological associations

Cancer occurred in 50 of 236 VGCC antibody seropositive neurological patients (21%, 46 had a history of cancer, and 6 had a new cancer found) and 34 of 202 seronegative controls (17%, all had a history of cancer only), $P = 0.27$. The neoplasms encountered among the patients are listed in Table S1 and are categorized according to antibody titer. Monoclonal gammopathy of undetermined significance and skin malignancies other than melanoma were excluded. One-fifth of the seropositive patients (50/236) had at least 1 neoplasm by history or detected following antibody detection (VGCC-P/Q antibody-positive only, 29; VGCC-N antibody-positive only, 15; dual antibody-positive, 6). A second cancer was found in 2 patients with past history of cancer.

Comprehensive cancer evaluation, performed in 123 patients [body computerized tomography (CT) scan, 105; positron-emission tomography (PET)-CT scan, 73] revealed imaging evidence of cancer in 6 patients (5%), a median of 3 months after autoantibody detection (range, 0–16 months), including small-cell lung carcinoma (2), breast adenocarcinoma (2), follicular lymphoma (1), and probable tonsillar carcinoma (1). One breast adenocarcinoma was discovered by routine diagnostic mammogram 13 months after the positive serology (body PET-CT scan had revealed no abnormality). The second patient's breast adenocarcinoma was identified by routine screening mammography 16 months after the positive serology (with negative initial mammogram). The patient with tonsillar

neoplasm suspected by PET-CT scan did not undergo recommended biopsy for confirmation and died within 2 months at another medical facility from undocumented cause.

Forty-six patients (19%) had a past history of 1 or more cancers of diverse histologic types and anatomic locations, including prostate adenocarcinoma (9), breast adenocarcinoma (8), cervical adenocarcinoma (4), lung small-cell carcinoma (3), bladder urothelial carcinoma (3), Hodgkin lymphoma (3), renal cell carcinoma (3), melanoma (3), colon adenocarcinoma (3), thyroid papillary carcinoma (2), non-Hodgkin lymphoma (1), lung squamous cell carcinoma (1), conjunctival spindle cell carcinoma (1), Waldenström macroglobulinemia (1), ovarian adenocarcinoma (1), vulval squamous cell carcinoma (1), gynecologic papillary serous adenocarcinoma (1), endometrial adenocarcinoma (1), lung adenocarcinoma (1), thymic carcinoma (1), multiple myeloma (1), adenocarcinoma of unknown primary (1), and myelofibrosis (1).

Neurological associations

Table 1 documents the frequency of neurological disorders. Definite or probable autoimmune diagnoses were assigned in 79 patients (35%) by the primary evaluating neurologist (53 and 26, respectively). In 6 patients the autoimmune disorder was multifocal, including LES (2, both had documented autonomic ganglionopathy in addition to the typical LES-associated dry mouth and constipation, and 1 had length-dependent somatic peripheral neuropathy), encephalomyelopathy (1), encephalomyeloradiculopathy (1), cerebellar ataxia and encephalopathy (1), and cerebellar ataxia with a prior history of myasthenia gravis (1). Forty-nine patients had neurological symptoms without objective findings and thus no neurological diagnosis. Thirteen patients were lost to follow up before final diagnostic classification. The following neurological disorders are described in rostro-caudal anatomic order. The comparative frequencies of neurological phenotypes among VGCC positive patients and 202 patient seronegative controls are documented in Supplementary Table S2. Dysautonomia, seizures, and neuromuscular junction disorders were more common among the VGCC antibody-positive patients.

Cortical disorders—Cognitive disorders (41) included autoimmune encephalopathy (14; 10 extra-limbic, 4 limbic), degenerative dementia (18), mild cognitive impairment (5), and cognitive disorder not otherwise specified (4). Ten patients had seizures in the setting of autoimmune encephalitis (4), autoimmune epilepsy (2), and seizures not otherwise specified (4).

Cerebellar ataxia—Nine of the 236 patients had a diagnosis of cerebellar ataxia (3.8%). We excluded from this category those patients whose ataxia was attributable to 1 or more radiologically-detectable cerebellar lesions consistent with stroke or classical multiple sclerosis. The median onset age was 59 years (range, 35–72 years); median follow up was 11 months (range, 1–48 months). Eight patients were VGCC-P/Q antibody-positive (median value, 0.10 nmol/L; range 0.07–4.16 nmol/L); 3 patients were VGCC-N antibody-positive (median value, 2.33 nmol/L, range, 0.05–6.08 nmol/L). Five patients in this group were assigned final diagnoses of autoimmune cerebellar ataxia, 3 patients with multiple system atrophy, cerebellar type (MSA-C, all had coexisting dysautonomia), and 1 with cerebellar

ataxia of unknown cause. Follow up information was available for 4 of 5 patients with final diagnoses of autoimmune cerebellar ataxia. Two had a paraneoplastic cause and died from cancer within a median of 5 months, and 2 improved with treatment.

Other movement disorders—Eleven patients had parkinsonism (all had been assigned a neurodegenerative diagnosis), 3 had dystonia (2 in the context of degenerative parkinsonism and 1 with isolated jaw and tongue dystonia of unknown cause), 1 had paraneoplastic chorea, and 1 had stiff-person syndrome.

Demyelinating central nervous system (CNS) disorders—Eight of 11 patients had a classical multiple sclerosis (MS) phenotype [primary progressive MS (5); relapsing remitting MS (3)], and 3 had an atypical demyelinating disorder [periventricular encephalitis (2), mono-hemispheric subcortical encephalitis with transverse myelitis (1)]. Four patients had a non-demyelinating myelopathy, 2 autoimmune and 2 of unknown cause.

Miscellaneous CNS disorders—Nine patients had a headache disorder, 9 had a degenerative spine disease, 9 had a toxic-metabolic neurologic disorder, 8 had cerebrovascular disease, and 2 had a brain neoplasm.

Anterior horn cell disorders—Eleven patients had been assigned the diagnosis of anterior horn cell disorder, 10 typical amyotrophic lateral sclerosis of neurodegenerative type and 1 phenotypically unusual with insidiously progressive bulbar involvement in the context of a history of Sjögren syndrome.

Peripheral neuropathies—Peripheral neuropathy was diagnosed in 61 patients (26%), and in 38 (16%) this was the only neurological finding (Table 1). In 22 of the 61 patients (36%), the cause was deemed autoimmune. Forty patients were VGCC-P/Q antibody-positive (median value, 0.07; range 0.04–4.16), and 28 were VGCC-N antibody-positive (median value, 0.06 nmol/L; range, 0.04–6.08).

Autonomic neuropathy or dysautonomia was encountered in 19 patients. Their final diagnoses were MSA (5), LES (2), postural orthostatic tachycardia syndrome (2), or other diagnosis (10).

Neuromuscular Junction Disorder—Neuromuscular junction disorders were documented in 10 of the 236 seropositive patients (4.2%). LES was confirmed electrophysiologically in 6 patients (median VGCC-P/Q antibody value, 0.49 nmol/L; range, 0.17–1.84 nmol/L). None was VGCC-N antibody-positive. A neoplasm (small-cell lung carcinoma) was identified prospectively after antibody detection in1 patient, a smoker aged 43 years.

Serological, clinical and electrophysiological evidence supported a postsynaptic neuromuscular transmission defect in another 4 patients (median VGCC-P/Q antibody value, 0.07 nmol/L; range 0.05–0.10; none was VGCC-N antibody-positive). Two had coexisting muscle AChR binding antibody; 2 had muscle AChR modulating Ab, and 1 had MuSK antibody, Table 2. Myasthenic findings were generalized in 3 and extraocular muscle-

restricted in 1. Tendon stretch reflexes were preserved in all. Compound muscle action potentials (CMAPs) were normal in all 3 who had nerve conduction testing. Responses to repetitive nerve stimulation were normal in 2 patients (1 with extraocular muscle-restricted weakness, the other with mild generalized weakness); 1 patient had a CMAP decrement (but not increment) and myopathic findings. Two of these 4 patients had a history of cancer (1 prostate adenocarcinoma and 1 metastatic gynecological serous adenocarcinoma).

Myopathies—Of 7 patients with diagnosis of myopathy, 3 had biopsy evidence of inflammation (varying patterns, but none diagnostically specific), 1 had necrotizing myopathy with minimal inflammatory reaction, and 3 were classified as non-autoimmune myopathy.

Clinical correlations according to antibody value

High values for VGCC-P/Q or VGCC-N antibody—Seven of the 236 patients had at least 1 VGCC antibody value exceeding 0.99 nmol/L, 3 of whom were positive for both antibodies. Five of those patients were assigned an autoimmune neurological diagnosis (71%, Tables S1 and 1): encephalopathy (3), LES (2), cerebellar ataxia (2), and peripheral neuropathy (2). One patient had a multifocal neurological disorder. Two patients had a history of cancer.

Medium values for VGCC-P/Q or VGCC-N antibody—Of the remaining 229 patients, 79 had at least 1 VGCC antibody value in the range of 0.10–0.99 nmol/L. Their assigned diagnoses were peripheral neuropathy (20), non-neurological disorder (16), cognitive disorder (9), autonomic neuropathy or dysautonomia (8), radiculopathy (7), neuromuscular junction disorder (5), myopathy (5), CNS demyelinating disease (5), myelopathy (4), parkinsonism (4), seizures (3), cerebellar ataxia (2), anterior horn cell disorder (2), brachial plexopathy (1), classic stiff-person syndrome (1), and miscellaneous neurological disorders (14). Nineteen of those patients (24%) had at least 1 autoimmune neurological diagnosis: encephalopathy or other cortical disorder (6), neuromuscular junction disorder (5), peripheral neuropathy (5), myopathy (3), or stiff-person syndrome (1). A malignancy was found in 4 patients following antibody detection.

Low values for VGCC-P/Q or VGCC-N antibody—In 150 patients the highest VGCC antibody value was in the range of 0.03–0.09 nmol/L. Their diagnoses included 1 or more of: peripheral neuropathy (39), non-neurological disorder (33), cognitive disorder (29), autonomic neuropathy or dysautonomia (11), anterior horn cell disorder (9), seizures (7), parkinsonism (7), CNS demyelinating disease (6), cerebellar ataxia (5), radiculopathy (3), brachial plexopathy (3), dystonia (3), neuromuscular junction disorder (3), myopathy (2), chorea (1), and miscellaneous CNS disorders (23). Twenty-nine patients (19%) had at least 1 autoimmune neurological diagnosis: peripheral neuropathy (15), encephalopathy or other cortical disorder (8), seizure disorder (5), neuromuscular junction disorder (3), ataxia (3), or another autoimmune neurological diagnosis (4). Cancer was found in 2 patients following antibody detection.

The overall positive predictive value for an autoimmune neurological diagnosis was 22%. Autoimmune neurological diagnoses were more common among patients with high antibody values (5/7) than among patients with medium values (19/79, P=0.02) or low values (29/150, P=0.004).

Coexisting autoantibodies

At least 1 other neural autoantibody was detected in 73 patients. Specificities included GAD65 (24), striated skeletal muscle (17), muscle AChR (binding antibody, 13; modulating antibody, 6), VGKC-complex (11), neuronal ganglionic AChR (10), AGNA-1 (1), NMDA receptor (1), $GABA_B$ receptor (1), ANNA-1 (1) and MuSK (1), Table 2. The GAD65 antibody value in 1 patient exceeded 20 nmol/L (3375 nmol/L); the neurological diagnosis was stiff-person syndrome. Non-organ-specific anti-nuclear antibody was noted in 19 patients, and smooth muscle antibody was noted in 1.

CSF Findings

CSF was tested in 75 patients. At least 1 abnormal value was recorded in 61 of the 75 patients (81%), most commonly an elevated protein (55 patients; median value of 58 mg/dL; range, 36–234). Thirteen had leukocytosis (median, 13 cells/μL; range, 6–43). Among those treated with immunotherapy, those with elevated CSF protein were not more likely to be responders (14 were responsive, 13 were not), but those with leukocytosis were responders (7 were responsive to immunotherapy, 0 were not).

Immunotherapies and outcomes among patients with suspected autoimmune diagnoses

Physician-documented symptomatic outcomes were available for 69 patients (29%) who received immunotherapy. Improvements were reported in 31 (mild to moderate, 22; robust, 9; Table 1). Some patients received more than 1 immunotherapy modality. Improvements were attributed to IV corticosteroids (10), oral corticosteroids (10), intravenous immune globulin (IVIg, 8), mycophenolate mofetil (4), azathioprine (2), plasma exchange (1), methotrexate (1), and rituximab (1).

DISCUSSION

We detected VGCC-P/Q or VGCC-N antibodies in 3.4% of patients who underwent comprehensive general serological evaluation for a neurological presentation which included neural-specific autoimmunity in the differential diagnosis. This study would have excluded most patients in whom LES was suspected clinically, because disease-specific testing can be ordered for LES. Previous reports have focused on LES, emphasizing lung carcinoma as the oncological association of VGCC antibodies, but VGCC-N and VGCC-P/Q antibodies were noted in an initial report in patients who presented with diverse paraneoplastic encephalomyeloneuropathies associated with adenocarcinomas of breast, ovary, colon, and other cancers.[6] The cancer types we encountered in the study reported here were more diverse (most frequently adenocarcinomas). These sequentially tested patients from Mayo Clinic neurology practice exhibited autoimmune neurological phenotypes extending to every level of the nervous system, and included, in addition to some cases of classical LES: encephalopathies, cerebellar ataxias, myelopathies, and inflammatory neuropathies. Because

frequency of cancer and most neurological phenotypes did not differ between cases and antibody negative controls, the significance VGCC-P/Q or VGCC-N antibodies must be interpreted in the context of other clinical and paraclinical findings.

We encountered diverse myasthenic syndromes, often paraneoplastic and reflecting the context in which testing was ordered. Six of 10 patients with a documented neuromuscular transmission defect unequivocally had LES, but 4 had clinical or electrophysiological findings that supported myasthenia gravis rather than LES. VGCC autoantibodies are rarely encountered in patients with myasthenia gravis except in paraneoplastic cases (other than thymoma).[11] Conversely, muscle AChR or striational antibodies are encountered in ~5–10% of patients with LES.[12–14]

As our laboratory has previously reported for the ganglionic α3 AChR and VGKC-complex antibodies, the neurological diagnostic specificity of seropositivity was directly proportional to the antibody level detected.[8,15] Patients with high values (≥ 1.00 nmol/L) usually had an autoimmune diagnosis. Those with medium or low values had autoimmune neurological diagnoses in 19–24%, but a further 20% had no neurological diagnoses after extensive clinical and paraclinical testing. For the remaining 60%, a neurological diagnosis of alternative cause was established (often neurodegenerative), or the medical records contained insufficient data to determine a definitive neurological diagnosis.

Because of these limitations, we advise that the neurological and oncological significance of VGCC-P/Q or VGCC-N antibody positivities be interpreted in the clinical context of the individual patient. Additional clues to an autoimmune diagnosis include a subacute onset disorder with rapid progression without obvious alternative diagnostic explanation, the presence of a classic phenotype (such as LES), a personal or family history of 1 or more coexisting autoimmune diseases, an inflammatory spinal fluid, an inflammatory appearing MRI scan of brain or spine, and a coexisting neural antibody.[16]

Thirty percent of patients had coexisting neural autoantibodies that in some instances enhanced the neurological diagnostic specificity, for example NMDA-R antibody detected in a patient with encephalitis, and GAD65 antibody detected in a patient with classical stiff-person syndrome.[17,18] Clues to an oncological diagnosis include a personal or family history of cancer and a smoking history. In other patients, the antibody profile had high specificity for cancer, for example ANNA-1 detected in a patient with small-cell carcinoma.[19]

In patients with isolated VGCC antibody findings with low suspicion for cancer based on the profile above, ensuring the patient is up-to-date (and continues to be) with age- and sex-appropriate cancer screening (breast exam, mammogram, pelvic exam, and colonoscopy) and a CT of chest (done 1 time only if normal) would likely suffice for cancer surveillance. A trial of immunotherapy could be considered in uncertain cases with objective neurological abnormalities, but it should not be undertaken in patients in whom an autoimmune diagnosis seems unlikely. False positivity may be encountered in patients with polyclonal hypergammaglobulinemia, including those who have received IVIg within the preceding 30 days.[8]

The VGKC-complex antibody radioimmunoassay, which is methodologically similar to the VGCC antibody assays employed in this study,[20,21] has been determined to detect autoantibodies which target neuronal proteins that co-immunoprecipitate with detergent-solubilized VGKCs (namely, Lgi1 and Caspr2).[22,23] Thus, it is conceivable that antibodies detected by the VGCC radioimmunoprecipitation assay might be directed at proteins interacting with VGCC-P/Q and VGCC-N proteins. A majority of patients with $GABA_BR$ antibodies have coexisting VGCC-P/Q or VGCC-N antibodies (77%).[24] In this study, only 1 patient with VGCC antibody had coexisting $GABA_BR$ autoantibody, indicating the rarity of $GABA_BR$ autoimmunity.

In summary, neuronal VGCC antibody-seropositive patients have diverse neurological presentations and cancer types. The cancers include squamous cell carcinomas, adenocarcinomas, and small-cell carcinomas. The neurological and oncological significance should be interpreted cautiously in the clinical context of the individual patient.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Abbreviations

AChR	acetylcholine receptor
AGNA	anti-glial/neuronal nuclear
AM	Andrew McKeon
AMPA	a-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid
ANNA	anti-neuronal nuclear
CASPR2	contactin-associated protein-2
CMAP	compound muscle action potential
CNS	central nervous system
CSF	cerebrospinal fluid
CRMP	collapsin response-mediator protein
GAD65	glutamic acid decarboxylase, 65 kDa isoform
GABA	gamma-aminobutyric acid
Ig	immune globulin
IVIg	intravenous immune globulin
LES	Lambert-Eaton syndrome
Lgi1	leucine-rich-glioma-inactivated-1

MS	multiple sclerosis
MSA	multiple system atrophy
MuSK	muscle-specific kinase
NMDA	n-methyl- D-aspartate
VGCC	voltage-gated calcium channel
VGKC	voltage-gated potassium channel

References

1. Fukunaga H, Engel AG, Lang B, Osame M, Lamber EH. Paucity and disorganization of presynaptic membrane active zones in the Lambert-Eaton myasthenic syndrome. Muscle Nerve. 1982; 5:686–697.
2. Roberts A, Perera S, Lang B, Vincent A, Newsom-Davis J. Paraneoplastic myasthenic syndrome IgG inhibits 45Ca2+ flux in a human small cell carcinoma line. Nature. 1985; 317:737–739. [PubMed: 2414666]
3. Lang B, Newsom-Davis J, Wray D, Vincent A, Murray N. Autoimmune aetiology for myasthenic (Eaton-Lambert) syndrome. Lancet. 1981; 2:224–226. [PubMed: 6114283]
4. De Aizpurua HJ, Lambert EH, Griesmann GE, Olivera BM, Lennon VA. Antagonism of voltage-gated calcium channels in small cell carcinomas of patients with and without Lambert-Eaton myasthenic syndrome by autoantibodies omega-conotoxin and adenosine. Cancer Res. 1988; 48:4719–4724. [PubMed: 2457429]
5. Sugiura Y, Woppmann A, Miljanich GP, Ko CP. A novel omega-conopeptide for the presynaptic localization of calcium channels at the mammalian neuromuscular junction. J Neurocytol. 1995; 24:15–27. [PubMed: 7539483]
6. Lennon VA, Kryzer TJ, Griesmann GE, O'Suilleabhain PE, Windebank AJ, Woppmann A, et al. Calcium-channel antibodies in the Lambert-Eaton syndrome and other paraneoplastic syndromes. N Engl J Med. 1995; 332:1467–1474. [PubMed: 7739683]
7. Motomura M, Johnston I, Lang B, Vincent A, Newsom-Davis J. An improved diagnostic assay for Lambert-Eaton myasthenic syndrome. J Neurol Neurosurg Psychiatry. 1995; 58:85–87. [PubMed: 7823075]
8. McKeon A, Lennon VA, Lachance DH, Fealey RD, Pittock SJ. Ganglionic acetylcholine receptor autoantibody: oncological, neurological, and serological accompaniments. Arch Neurol. 2009; 66:735–741. [PubMed: 19506133]
9. McKeon A, Lennon VA, LaChance DH, Klein CJ, Pittock SJ. Striational antibodies in a paraneoplastic context. Muscle Nerve. 2013; 47:585–587. [PubMed: 23463255]
10. Apiwattanakul M, McKeon A, Pittock SJ, Kryzer TJ, Lennon VA. Eliminating false-positive results in serum tests for neuromuscular autoimmunity. Muscle Nerve. 2010; 41:702–704. [PubMed: 20405502]
11. Vernino S, Lennon VA. Autoantibody profiles and neurological correlations of thymoma. Clin Cancer Res. 2004; 10:7270–7275. [PubMed: 15534101]
12. Lennon VA. Serological profile of myasthenia gravis and distinction from the Lambert-Eaton myasthenic syndrome. Neurology. 1997; 48:S23–S27.
13. Tim RW, Massey JM, Sanders DB. Lambert-Eaton myasthenic syndrome (LEMS). Clinical and electrodiagnostic features and response to therapy in 59 patients. Annals of the New York Academy of Sciences. 1998; 841:823–826. [PubMed: 9668336]
14. Leys K, Lang B, Vincent A, Newsom-Davis J. Calcium channel autoantibodies in Lambert-Eaton myasthenic syndrome. Lancet. 1989; 2:1107. [PubMed: 2572837]

15. Klein CJ, Lennon VA, Aston PA, McKeon A, O'Toole O, Quek A, et al. Insights from LGI1 and CASPR2 potassium channel complex autoantibody subtyping. JAMA Neurol. 2013; 70:229–234. [PubMed: 23407760]
16. McKeon A, Lennon VA, Pittock SJ. Immunotherapy-responsive dementias and encephalopathies. Continuum (Minneap Minn). 2010; 16:80–101. [PubMed: 22810282]
17. McKeon A, Robinson MT, McEvoy KM, Matsumoto JY, Lennon VA, Ahlskog JE, et al. Stiff-man syndrome and variants: clinical course, treatments, and outcomes. Arch Neurol. 2012; 69:230–238. [PubMed: 22332190]
18. Kruse JL, Lapid MI, Lennon VA, Klein CJ, Toole OO, Pittock SJ, et al. Psychiatric Autoimmunity: N-Methyl-D-Aspartate Receptor IgG and Beyond. Psychosomatics. 2015; 56:227–241. [PubMed: 25975857]
19. Horta ES, Lennon VA, Lachance DH, Jenkins SM, Smith CY, McKeon A, et al. Neural autoantibody clusters aid diagnosis of cancer. Clin Cancer Res. 2014; 20:3862–3869. [PubMed: 24833664]
20. Hart IK, Waters C, Vincent A, Newland C, Beeson D, Pongs O, et al. Autoantibodies detected to expressed K+ channels are implicated in neuromyotonia. Ann Neurol. 1997; 41:238–246. [PubMed: 9029073]
21. Vernino S, Lennon VA. Ion channel and striational antibodies define a continuum of autoimmune neuromuscular hyperexcitability. Muscle Nerve. 2002; 26:702–707. [PubMed: 12402293]
22. Irani SR, Alexander S, Waters P, Kleopa KA, Pettingill P, Zuliani L, et al. Antibodies to Kv1 potassium channel-complex proteins leucine-rich, glioma inactivated 1 protein and contactin-associated protein-2 in limbic encephalitis, Morvan's syndrome and acquired neuromyotonia. Brain. 2010; 133:2734–2748. [PubMed: 20663977]
23. Lai M, Huijbers MG, Lancaster E, Graus F, Bataller L, Balice-Gordon R, et al. Investigation of LGI1 as the antigen in limbic encephalitis previously attributed to potassium channels: a case series. Lancet Neurol. 2010; 9:776–785. [PubMed: 20580615]
24. Jeffery OJ, Lennon VA, Pittock SJ, Gregory JK, Britton JW, McKeon A. GABAB receptor autoantibody frequency in service serologic evaluation. Neurology. 2013; 81:882–887. [PubMed: 23925760]
25. O'Toole O, Lennon VA, Ahlskog JE, Matsumoto JY, Pittock SJ, Bower J, et al. Autoimmune chorea in adults. Neurology. 2013; 80:1133–1144. [PubMed: 23427325]

Table 1

Neurological Findings & Immunotherapy Responses

Neurological disorder	Total patients (N=236)	Classified according to highest VGCC-P/Q or VGCC-N Ab value			Both VGCC-P/Q and VGCC-N detected (N=21)	Immunotherapy response		
		High (N=7)	Medium (N=79)	Low (N=150)		No response (N=38)	Modest response (N=22)	Robust Response (N=9)
Peripheral neuropathy *	61	2	20	39	9	6	4	2
None	49	0	16	33	0	1	0	1
Cognitive	41	3	9	29	5	8	0	3
Miscellaneous	37	0	14	23	1	3	0	0
Dysautonomia	19	0	8	11	0	3	2	0
Parkinsonism	11	0	4	7	1	2	0	0
Demyelinating disease	11	0	5	6	2	1	1	1
Anterior horn cell disorders	11	0	2	9	1	3	0	0
Radiculopathy	10	0	7	3	2	2	0	0
Seizures	10	0	3	7	0	1	0	1
Neuromuscular junction defect	10	2	5	3	0	0	8	0
Cerebellar ataxia (non-demyelinating)	9	2	2	5	2	3	2	0
Myopathy	7	0	5	2	0	1	3	0
Brachial plexopathy	4	0	1	3	0	0	1	1
Myelopathy (non-demyelinating)	4	0	4	0	0	2	0	0
Dystonia	3	0	0	3	0	0	0	0
Chorea	1	0	0	1	0	1	0	0
Sensorineural hearing loss	1	0	0	1	0	1	0	0
Stiff person syndrome	1	0	1	0	0	0	1	0

* EMG available in 55: length-dependent and primarily axonal sensorimotor (33), length-dependent mixed axonal and demyelinating (4), brachial plexopathy (4), sensorimotor demyelinating (2), sensory neuronopathy (2), severe, diffuse motor-predominant axonal polyradiculoneuropathy (1), multiple mononeuropathies (1), multiple subacute cranial neuropathies (1), multiple chronic cranial neuropathies (1), mild axonal neuropathy with fasciculations (1), severe chronic diffuse patchy polyradiculoneuropathy (1), sensorimotor polyradiculoneuropathy with demyelinating features (1), patchy primarily axonal polyradiculoneuropathy (1), and normal findings (2). Three patients were diagnosed with small fiber neuropathy only (normal EMG, 2; on clinical grounds alone, 1). Biopsied sural nerve tissue (2 patients) did not reveal inflammation.

Abbreviations: VGCC, voltage-gated calcium channel.

Table 2

Coexisting neural autoantibodies

Antibody	Patients (N=73)	Median value	Range of positive values	Normal range	Neurological and cancer diagnoses
GAD65	24	0.15 nmol/L	0.03–3375 nmol/L	≤ 0.02 nmol/L	Various
Striated muscle	17	1: 480	1:120–1:61440	< 1: 120	Various
AChR binding	13	0.12 nmol/L	0.03–28.0 nmol/L	≤ 0.02 nmol/L	Various
VGKC-complex	11*	0.17 nmol/L	0.03–1.74 nmol/L	≤ 0.02 nmol/L	Various
Ganglionic AChR	10	0.07 nmol/L	0.03–0.20 nmol/L	≤ 0.02 nmol/L	Various
AChR modulating	6	45% loss	28–94% loss	≤ 20% loss	Various
ANNA-1	1	1: 122,880	-	≤ 1: 120	Sensory neuronopathy, small-cell carcinoma
AGNA-1	1	1:1920	-	≤ 1: 120	LES, small-cell carcinoma
NMDA receptor	1	CSF positive (CBA)	-	-	Encephalitis, none
GABAB receptor	1	Serum positive (CBA; CSF not available)	-	-	Chronic whole-body pain (nos); malignant thymoma
MuSK	1	5.13 nmol/L	-	≤ 0.02 nmol/L	Myasthenia gravis; none

*One patient each was seropositive for Lgi1 and CASPR2 antibodies. The former had autoimmune encephalitis, the latter had autoimmune chorea.[25]

Abbreviations: AChR, acetylcholine receptor; AGNA, antiglial/neuronal nuclear antibody; ANNA, antineuronal nuclear antibody; CBA, cell-based assay; CSF, cerebrospinal fluid; GABA, gamma-aminobutyric acid; GAD65, glutamic acid decarboxylase, 65 kD isoform; MuSK, muscle-specific kinase; NMDA, n-methyl-D-aspartate; VGKC, voltage-gated potassium channel complex.