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Clinical Study

When should we test for voltage-gated potassium channel complex antibodies? A retrospective case control study

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ABSTRACT

Patients with voltage-gated potassium channel (VGKC)-complex antibodies are increasingly recognized as having central, peripheral or combined phenotypes. With increasing awareness, more patients are tested and the clinical spectrum is expanding. Consequently, clinicians may be uncertain as to which patients should or should not be tested. Previous studies have identified common clinical features, but none has looked at the usefulness of these in predicting seropositive disease. We conducted a case-control study of patients tested for VGKC-complex antibodies over 10 years at a regional tertiary neurology centre determining which clinical/biochemical features were associated with antibody-positive disease. We found a marked increase in the numbers tested, although the percentage positive remained low. Antibody titre was highest in central disease ($p < 0.001$). Time from presentation to testing was shorter in those with VGKC-disease ($p = 0.01$). Seizures were present in 11 (69%) of those with VGKC-disease versus three (18%) without (odds ratio [OR] 10.3, 95% confidence interval [CI]: 2.0–52.7, $p = 0.005$). There was an inverse correlation between the antibody titre and serum sodium. A multivariate model selected seizures and hyponatraemia as predictive of VGKC disease (sensitivity 75% and specificity 82%); faciobrachial dystonic movements were specific but insensitive. Interestingly serum alkaline phosphatase was higher in those with VGKC-disease ($p = 0.016$) and highest in those with peripheral disease ($p = 0.015$). An ALP > 70 u/L was strongly associated with antibody positivity (OR 4.11 95% CI: 1.43–11.8, $p = 0.007$) with a sensitivity of 74.2%. The presence of seizures, faciobrachial movements, and hyponatraemia should raise suspicion of VGKC-disease; alkaline phosphatase may represent a novel biomarker, particularly in those with peripheral disease.

1. Introduction

Voltage-gated potassium channel (VGKC)-complex antibodies were initially identified in association with peripheral nerve hyper-excitability [1–5]. They are now recognised to be associated with clinical syndromes of both the peripheral (PNS) and central (CNS) nervous system, causing neuromyotonia and limbic encephalitis respectively, or a combined phenotype termed Morvan's syndrome [6–9]. This variation in clinical phenotypes is attributable to different antibody targets on the extracellular domains of neuronal cell membrane proteins, with antibodies directed against contactin-2 associated proteins (CASPR2) causing predominantly peripheral disease and those against leucine-rich glioma inactivated 1 (LGI1) associated with central manifestations [10–14]. Regardless of clinical presentation, early intervention is pivotal, particularly as timely immunotherapy can significantly improve outcome in those with CNS disease [15–19].

However, an increasingly diverse clinical picture of VGKC disease is emerging. Several studies have implicated VGKC-complex antibodies in epilepsy, chronic pain, neuropsychiatric presentations and disorders of movement and autonomic function [20–24]. An expanding clinical spectrum, growing awareness and improved availability of serum assays has led to an increase in VGKC-complex antibody testing [25]. However, a positive serum

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VGKC-complex antibody assay does not preclude an alternative diagnosis [22,26–29].

This presents a challenge for neurologists considering testing for VGKC-complex antibodies, as excessive testing may be unnecessarily costly and potentially misleading, whilst not testing can lead to missed diagnoses. Therefore, we analysed the clinical and biochemical features of patients in whom VGKC-complex antibody testing had been performed to identify the clinical and laboratory features associated with antibody-positive disease.

2. Methods

2.1. Patients

We conducted a retrospective case control study at the Walton Centre NHS Foundation Trust, a regional tertiary neurology centre in the UK, serving a population of 3.5 million people. The electronic laboratory database was screened over a 10-year period (1st April 2001 to 1st July 2011), to identify adults (>16 years) whose serum had been tested for VGKC-complex antibodies. Before 1st May 2005 sample processing was outsourced to a second laboratory; after this date all samples were processed internally [29]. A positive titre was defined as $\geqslant$100 pmol/L on the Oxford assay and $\geqslant$85 pmol/L on the commercial assay used at the Walton Centre. Because the number of negative patients was many more than positives, and we wanted to analyse similar numbers, we randomly selected negative patients (1:1) with a random number generator [30].

Both paper and computer-based clinical case notes were retrieved and examined by members of the team (AD, BO, EK, TS). Data were collected using a standardised proforma for demographics, clinical features, investigation results at the time of testing and the subsequent treatment, informed by syndromes of VGKC-complex antibody-associated disease [6–8,20,22].

To reduce measurement bias, the data collection proforma was refined after an initial analysis of a subset of patients that was performed in duplicate by two members of the team (BO, BDM). Study methodology and analysis are presented in accordance with STrengthening the Reporting of OBservational studies in Epidemiology (STROBE) guidelines [31].

2.2. Case definitions

Antibody-positive cases were classified into clinical case definitions according to clinical features and VGKC-complex antibody titre as having CNS disease, PNS disease, both CNS/PNS disease, or as clinically-insignificant positive (Table 1). A case was defined as a clinically-insignificant positive if a positive VGKC-complex antibody titre was found, in the presence of a more likely alternative diagnosis, for example clinical features, electrophysiology and positive acetylcholine receptor antibody titres in keeping with myasthenia gravis. Where cases met one or more of these definitions, the most likely diagnosis as judged clinically was used, and any disagreements resolved through discussion with a more senior author (BDM, BO, TS). A case series of a subset of patients has been presented but not as a formal case-control study [29].

2.3. Statistical methods

Univariate statistical analysis was conducted using PRISM (GraphPad PRISM 2014). Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) of clinical features were calculated and odds ratios with 95% confidence intervals generated. Chi-squared or Fisher's exact tests were used to compare categorical data and the Mann–Whitney *U* test was used for continuous, non-parametric data. To minimise the potential for type I error, clinical features occurring in less than 10% of VGKC-complex antibody positive patients were excluded from the analysis. Hyponatraemia was defined by local laboratory protocol as sodium <133 mmol/L. The normal reference range for alkaline phosphatase (ALP) was 30–130 units/L).

Multivariate logistic regression analysis was performed using SPSS (SPSS v20 2012). The presence of VGKC-complex antibody-associated disease was used as a binary outcome measure. In order that missing data did not bias the multivariate analysis, variables that were not recorded in the majority of patients were excluded. Statistical significance was defined as $P < 0.05$.

Table 1
Clinical case definitions of patients tested for voltage-gated potassium channel (VGKC)-complex antibodies in a tertiary care centre

Serum antibody titre	Clinical case definition	Criteria
Positive titre*	VGKC-complex antibody-associated CNS disease (Limbic Encephalitis)	Seizures and/or neuropsychiatric symptoms. Absence of peripheral motor or sensory signs and symptoms
	VGKC-complex antibody-associated PNS disease (Isaac's syndrome/ Neuromyotonia)	Clinical evidence of peripheral nerve hyperexcitability. Absence of central nervous signs and symptoms.
	VGKC-complex antibody-associated CNS and PNS disease (Morvan's syndrome)	Seizures and/or neuropsychiatric symptoms and clinical evidence of peripheral nerve hyperexcitability
	Clinically-insignificant positive	Alternative diagnosis more likely
Negative titre*	VGKC-complex antibody negative disease	

* A positive titre was defined as $\geqslant$100 pmol/L on the Oxford assay and $\geqslant$85 pmol/L on the commercial assay at the Walton Centre.
CNS = central nervous system, PNS = peripheral nervous system.

3. Results

3.1. Identification and clinical diagnosis

Between 2001 and 2011, 1236 patients were tested for VGKC-complex antibodies, of whom 87 (7%) were positive (Fig. 1). Over this period, we identified a large increase in the numbers of patients tested. Between the first and second quarter studied the median (range) number tested per month rose from 3.5 (1–7) to 7 (3–13), $p < 0.0001$; and between the second and third quarter studied this rose from 7 (3–13) to 15 (7–26), $p < 0.0001$. Although there was an increase in the number tested per month between the third to fourth quarters studied this was not significant, 15 (7–26) to 16 (9–27), $p = 0.045$. Despite this increased testing over these periods, there was no significant increase in the number of positive patients identified, and the percentage of positive patients remained low in each quarter studied (3.2%, 9.3%, 5.2%, and 7.0%, respectively), and the percentage of those with VGKC-complex disease was low throughout the study period (Fig. 2). Clinical information was available for 62 (71%) of patients with a positive antibody titre. Of these 35 (56%) met the clinical case definition for VGKC-complex disease; 13 (37%) had CNS disease, 17 (49%) had PNS disease and 5 (14%) had combined CNS/PNS disease. Twenty-seven (44%) met the clinical case definition for false positive and had a broad range of final diagnoses, many of which were other autoimmune conditions (Table 2a). In 24 (85%) false positive cases there were clear neuroimaging, neurophysiological, and/or