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No evidence for genetic association between glutamate transporter EAAT2 and Devic's neuromyelitis optica in caucasians and afro-caribbeans

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Abstract
Devic's neuromyelitis optica (NMO) is a severe inflammatory and autoimmune disease producing demyelinating lesions. Recent data suggest that a complex genetic component could be involved. While impairment of glutamate homeostasis has emerged as a contributing etiological factor in NMO, a genetic alteration of Excitatory Amino Acid Transporter 2 (*EAAT2*/*SLC1A2*), the major glutamate transporter in the Central Nervous System (CNS), could contribute to glutamate excitotoxicity and then must be considered.
We evaluated whether mutations and/or single nucleotide polymorphisms (SNPs) in *EAAT2* gene, are associated with susceptibility to NMO. We studied a cohort of NMO sporadic cases including afro-caribbean patients ($n=81$; French cohort of Devic's neuromyelitis optica—NOMADMUS cohort) and compared to control subjects ($n=56$). We sequenced the whole coding region of *EAAT2* gene and splicing consensus sequences flanking each exon. The results obtained from all NMO samples did not show any novel mutations and/or SNPs both in the coding region and splicing sites of *EAAT2* gene compared to controls subjects. We reported three synonymous SNPs (rs752949, rs1042113 and rs7102949) but only rs7102949 was found in afro-caribbean. Genotype frequencies did not differ between patients and controls for the three SNPs in caucasians and afro-caribbeans (rs752949: $p=0.71$ and $p=0.37$, respectively; rs1042113: $p=0.73$ and $p=0.35$, respectively; rs7102949: $p=0.08$ in afro-caribbeans). Our data showed no evidence for a genetic association between *EAAT2* gene and Devic's neuromyelitis optica.

1. Introduction

Neuromyelitis optica (Devic's disease, NMO) is a rare and severe inflammatory and demyelinating disease characterized by attacks of myelitis and optic neuritis (Wingerchuk et al., 2007). In addition to these clinical data necessary for diagnostic criteria, a specific antibody called NMO-IgG, which bind to the dominant central nervous system (CNS) water channel protein Aquaporin 4 (AQP4) was identified, and then has been included in the new diagnostic criteria for NMO since 2006 (Lennon et al., 2004; Wingerchuk et al., 2006).

Recently, it has been demonstrated that the frequency of familial NMO cases was higher compared to the NMO prevalence found in the general population highlighting the involvement of a complex genetic component in susceptibility to NMO (Matiello et al., 2010). Genetic analysis demonstrated alterations in *HLA class II DRB1 03* gene, cytochrome P450 *CYP7A1* gene, and *AQP4* gene in NMO (Deschamps et al., 2010; Kim et al., 2009; Matiello et al., 2011).

Excitatory amino acid transporter 2 (*EAAT2*/*SLC1A2*) is expressed by astrocytes, and plays a major role in glutamate homeostasis. Its activity was reported to be affected by NMO-IgG and AQP4 (Hinson et al., 2008). As impairment of glutamate homeostasis has emerged as a contributing etiological factor in CNS disease including NMO (Hinson et al., 2008; Marignier et al., 2010), astrocytic glutamate uptake could be directly affected by mutations or polymorphisms in *EAAT2* gene.

Our study aims to examine the involvement of *EAAT2* in NMO pathogenesis. Our primary objective was to screen the EAAT2 gene for mutations in the whole coding region of NMO patients coming from the French cohort of Devic's neuromyelitis optica (NOMADMUS cohort) and an afro-caribbean cohort. Our secondary objective was to analyze the putative role of mutations/SNPs in those patients.

2. Materials and methods

2.1. Patients

We studied 81 NMO cases fulfilling the 2006 criteria for NMO (Wingerchuk et al., 2006): 50 caucasians (12 males and 38 females; mean age: 45.6 ± 11.8 years old; 27 patients positive for anti-AQP4 antibody) and 31 french afro-caribbeans (three males and 28 females; mean age: 47.1 ± 14.5 years old). DNA samples of NMO patients were obtained from the French cohort of Devic's neuromyelitis optica (NOMADMUS cohort, Neurobiotec, Biological Resource Center of Hospices civils de Lyon) and from the Afro-Caribbean cohort (CeRBiM, Biological Resource Center of Martinique).

Fifty six controls were also studied: 26 caucasians aged over 50 years with no signs of neurodegenerative disorder

(*footnote continued*)
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