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LETTER TO THE EDITOR

Reply: *In vitro* effects of a human monoclonal antibody against the *N*-methyl-D-aspartate receptor

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Sir,

We thank Professor Dalmau for the stimulating thoughts on our work (Dalmau, 2016). We fully agree that—already before our study—the fundamental experimental data acquired by his and further groups using human immunoglobulins from patients with *N*-methyl-D-aspartate receptor (NMDAR) encephalitis provided solid evidence that the clinical symptoms relate to dysfunction of NMDA receptors caused by the antibodies contained in human samples (Dalmau, 2008; Hughes *et al.*, 2010; Prüss *et al.*, 2010; Moscato *et al.*, 2014; Planaguma *et al.*, 2015, 2016). Effects in primary neuronal cell cultures mimicked the phenotype in laboratory animals and were thus designated as surrogate markers of the pathophysiology. Therefore, in this first paper using monoclonal human NMDAR autoantibodies, we focused on the established surrogate effects. Ongoing work with *in vivo* models will eventually confirm whether similar effects result from these (and only these) antibodies. In addition to this, the scientific community is most welcome to use these antibodies in their areas of expertise, ranging from high-resolution imaging to animal models to synaptic remodelling. In our opinion, the main achievement of the present work is not recapitulating antibody effects, but rather showing that monoclonal antibodies can be cloned from encephalitis patients and be used for biochemical and electrophysiological assays in exactly determined concentrations of the disease-specific immunoglobulin, which is impossible with human samples. We feel that this increase in availability and standardization will become a gold standard in future papers on human encephalitis autoantibodies. More importantly, precisely determined concentrations, known epitopes and antibody affinities, reproducible conditions between laboratories and many more are advantages of disease-specific monoclonal antibodies and will eventually also solve the controversial literature on the relevance of non-IgG isotype antibodies (Prüss *et al.*, 2012; Dahm *et al.*, 2014; Doss *et al.*, 2014; Dalmau, 2016).

We respectfully disagree that co-existence of other antibodies is rare. As outlined in detail in the paper, we found in all patients live plasma cells that produced antibodies against further, mainly not yet specified brain epitopes. Plasma cells are continuously releasing antibodies at the rate of several thousand immunoglobulin molecules per second (Alberts *et al.*, 2002). We agree, though, that this amount might not reach the detection threshold in clinical routine assays. In turn, low antibody levels certainly do not exclude a participation in the disease process. Unfortunately, immunoadsorption of patients' CSF with HEK cells expressing the NMDAR (Planaguma *et al.*, 2016) cannot unambiguously exclude the participation of further low-level antibodies. Here, ~10% of our monoclonal non-NR1 antibodies strongly bound to HEK cells and would therefore be similarly removed.

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Interestingly, our ongoing attempts to identify the target epitopes of the non-NR1 monoclonal antibodies, using immunoprecipitation and mass spectrometry, resulted in attractive first candidates, such as ryanodine receptor 2 (RYR2). RYR2 antibodies are not only discussed as being pathogenic in humans (Gilhus *et al.*, 2016), genetic dysfunction of RYR2 results in fatal cardiac arrhythmias and asystole (Meli *et al.*, 2011). Indeed, the young NMDAR encephalitis patient from whom we cloned this antibody suffered from repeated asystole and required a transient pacemaker during her intensive care unit stay. We feel that the most interesting question emerging from this controversy is not whether further antibodies are present, but why and when some of these antibodies (AQP4, MOG, AChR, GFAP) rise to detectable levels and lead to overt symptoms (Leite *et al.*, 2012; Titulaer *et al.*, 2014; Fang *et al.*, 2016), then classified as co-existing autoimmune entities, such as NMDAR encephalitis, neuromyelitis optica (NMO)-spectrum disorder or myasthenia gravis.

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References

Alberts B, Johnson A, Lewis J, Raff M, Roberts K, Walter P. Molecular biology of the cell. . New York, NY: Garland Science; 2002.

Dahm L, Ott C, Steiner J, Stepniak B, Teegen B, Saschenbrecker S, et al. Seroprevalence of autoantibodies against brain antigens in health and disease. Ann Neurol 2014; 76: 82–94.

Dalmau J. *In vitro* effects of a human monoclonal antibody against the N-methyl-d-aspartate receptor (Letter to the Editor). Brain 2016, in press.

Dalmau J, Gleichman AJ, Hughes EG, Rossi JE, Peng X, Lai M, et al. Anti-NMDA-receptor encephalitis: case series and analysis of the effects of antibodies. Lancet Neurol 2008; 7: 1091–8.

Doss S, Wandinger KP, Hyman BT, Panzer JA, Synofzik M, Dickerson B, et al. High prevalence of NMDA receptor IgA/IgM antibodies in different dementia types. Ann Clin Transl Neurol 2014; 1: 822–32.

Fang B, McKeon A, Hinson SR, Kryzer TJ, Pittock SJ, Aksamit AJ, et al. Autoimmune Glial Fibrillary Acidic Protein Astrocytopathy: A Novel Meningoencephalomyelitis. JAMA Neurol 2016; doi: 10.1001/jamaneurol.2016.2549. [Epub ahead of print]

Gilhus NE, Skeie GO, Romi F, Lazaridis K, Zisimopoulou P, Tzartos S. Myasthenia gravis - autoantibody characteristics and their implications for therapy. Nat Rev Neurol 2016; 12: 259–68.

Hughes EG, Peng X, Gleichman AJ, Lai M, Zhou L, Tsou R, et al. Cellular and synaptic mechanisms of anti-NMDA receptor encephalitis. J Neurosci 2010; 30: 5866–75.

Leite MI, Coutinho E, Lana-Peixoto M, Apostolos S, Waters P, Sato D, et al. Myasthenia gravis and neuromyelitis optica spectrum disorder: a multicenter study of 16 patients. Neurology 2012; 78: 1601–7.

Meli AC, Refaat MM, Dura M, Reiken S, Wronska A, Wojciak J, et al. A novel ryanodine receptor mutation linked to sudden death increases sensitivity to cytosolic calcium. Circ Res 2011; 109: 281–90.

Moscato EH, Peng X, Jain A, Parsons TD, Dalmau J, Balice-Gordon RJ. Acute mechanisms underlying antibody effects in anti-N-methyl-D-aspartate receptor encephalitis. Ann Neurol 2014; 76: 108–19.

Planaguma J, Haselmann H, Mannara F, Petit-Pedrol M, Grunewald B, Aguilar E, et al. Ephrin-B2 prevents N-methyl-D-aspartate receptor antibody effects on memory and neuroplasticity. Ann Neurol 2016; 80: 388–400.

Planaguma J, Leypoldt F, Mannara F, Gutierrez-Cuesta J, Martin-Garcia E, Aguilar E, et al. Human N-methyl D-aspartate receptor antibodies alter memory and behaviour in mice. Brain 2015; 138: 94–109.

Prüss H, Dalmau J, Harms L, Höltje M, Ahnert-Hilger G, Borowski K, et al. Retrospective analysis of NMDA receptor antibodies in encephalitis of unknown origin. Neurology 2010; 75: 1735–9.

Prüss H, Höltje M, Maier N, Gomez A, Buchert R, Harms L, et al. IgA NMDA receptor antibodies are markers of synaptic immunity in slow cognitive impairment. Neurology 2012; 78: 1743–53.

Titulaer MJ, Höftberger R, Iizuka T, Leypoldt F, McCracken L, Cellucci T, et al. Overlapping demyelinating syndromes and anti–N-methyl-D-aspartate receptor encephalitis. Ann Neurol 2014; 75: 411–28.

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