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Autoantibodies against *N*-methyl-*d*-aspartate receptor 1 in health and disease

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Purpose of review
Humoral autoimmunity has gained highest interest in neurology and psychiatry. Despite numerous recent articles on this hot topic, however, the biological significance of natural autoantibodies (AB) and the normal autoimmune repertoire of mammals remained quite obscure. AB may contribute to disorder-relevant phenotypes and are even believed to induce diseases themselves, but the circumstances under which AB become pathogenic are not fully understood. This review will focus on the highly frequent AB against the *N*-methyl-*d*-aspartate receptor 1 (NMDAR1-AB) as an illustrating example and provide a critical overview of current work *(please note that the new nomenclature, GluN1, is disregarded here for consistency with the AB literature*). In particular, it will demonstrate how little is known at this point and how many conclusions are drawn based on small numbers of individuals, fragmentary experimental approaches or missing controls.

Recent findings
NMDAR1-AB were investigated by clinicians world-wide with numerous small studies and case reports appearing yearly. Many publications were on 'anti-NMDAR encephalitis' cases or tried to separate those from other NMDAR1-AB associated conditions. Original exclusivity claims (e.g. electroencephalogram, EEG or functional magnetic resonance imaging, fMRI findings) turned out not to be exclusive for 'anti-NMDAR encephalitis'. Systematic analyses of representative NMDAR1-AB positive sera of all immunoglobulin (Ig) classes showed comparable distribution of different epitopes, often polyspecific/polyclonal, across health and disease. Sophisticated imaging tools provided findings on synapse trafficking changes induced by NMDAR1-AB from psychotic subjects but still lack epitope data to support any claimed disorder link. Persistently high titers of NMDAR1-AB (IgG) in immunized mice with open blood–brain barrier (BBB)-induced psychosis-like symptoms but failed to induce inflammation in the brain. Knowledge on peripheral NMDAR, for example in the immune system, and on potential inducers of NMDAR1-AB is only slowly increasing.

Summary
The present knowledge on the (patho) physiological role of NMDAR1-AB is very limited and still characterized by adamant rumors. Much more experimental work and more solid and informative clinical reports, including large numbers of subjects and adequate control groups, follow-up investigations and interdisciplinary approaches will be necessary to obtain a better understanding of the significance of humoral autoimmunity in general *(in focus here: NMDAR1-AB)* and its disease-relevance in particular.

Keywords
antigen, anti-*N*-methyl-*d*-aspartate receptor encephalitis, B cell, brain, epitope, functionality, humoral, immunoglobulin class, psychosis, stress

INTRODUCTION

The biological significance of natural AB and the normal autoimmune repertoire of mammals – even though principally recognized for many decades – has to a large degree remained mysterious. Natural AB of different classes are produced from early development on in the absence of exposure to foreign antigens. Their physiological role is only partly understood, and little is known about the circumstances and mechanisms that may potentially turn these natural 'harmless' AB of the normal autoimmune repertoire into symptomatic or disease-relevant AB.

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KEY POINTS

News, pitfalls and suggestions for improvement

- Functional NMDAR1-AB of all classes are highly seroprevalent across mammals, health and disease.
- Different epitopes recognized by NMDAR1-AB were identified, located in the extracellular part of NMDAR1, intracellular, C-terminal and in the extralarge pore domain. No consistent epitope pattern was detectable regarding immunoglobulin class or health/disease state. Half of the screened sera showed polyclonality/polyspecificity.
- 'Anti-NMDAR encephalitis' is not a distinct disease entity but probably comprises any brain inflammation where NMDAR1-AB happen to be present and syndrome-shaping. NMDAR1-AB (IgG) do not cause encephalitis (inflammation) on their own.
- All NMDAR1-AB can upon access to the brain – via leaky BBB or intrathecal production – contribute to neuropsychiatric symptoms in absence of any inflammation.
- To advance the AB research field, more comprehensive information should be requested for publications, e.g. solid description of methods, Ig classes, titers etc.
- Improvement of methods for NMDAR1-AB (and other AB) determination should be mandatory: ELISA is insufficient; functionality testing should be integrated.
- Claims regarding any disease relevance require appropriate controls, larger numbers of subjects and information on BBB function.

According to the present concept, natural AB (IgM, IgA or IgG) are made by B1 cells and marginal zone splenic B cells, independent of T cell help. In contrast, immune IgM and subsequently IgG or IgA are produced in response to foreign antigens by B2 cells that require antigen binding to the B-cell receptor and additional T-cell support to generate AB. The IgM secreting B2 cells migrate to B-cell follicles where, with the help of T cells, they undergo isotype switching and somatic hypermutation. Resulting are long-lived memory B cells, capable of differentiating into plasma cells that produce high-affinity binding IgG or IgA [1–4]. Whether, where and how these two general models intermingle, leading to autoimmune disease, is still quite nebulous.

Based on protein arrays and immunostaining of brain sections, human subjects as well as other mammals were reported to carry up to 1000 different serum AB, many directed against yet to be identified brain antigens, with a pattern highly variable among individuals [5,6]. This astonishing finding indicates that these AB might be important mediators, for instance in regulating immune responses, including immune maturation and tolerance induction, dampening inflammation, or in apoptotic cell clearance and debris removal [1,2,6]. Of potential diagnostic relevance is their new application as biomarkers revealing or excluding specific disease signatures [7,8▪,9,10].

The substantially growing interest in brain antigen-directed AB is reflected by numerous recent reviews (e.g. [11▪,12–14,15▪,16,17▪]). The search for their impact on neuropsychiatric disorders has led to new unexpected findings that question hitherto existing dogmas. AB previously believed to be illness indicators turned out to be equally detectable in healthy individuals. Screening of > 4200 individuals revealed an identical seroprevalence of 25 brain antigen-directed AB across various different disease groups and healthy subjects. Also, distribution of immunoglobulin classes (IgM, IgG and IgA) and AB titers turned out comparable, thus challenging an unambiguous causal relationship of any of these AB with brain disease [18]. A somewhat perplexing observation in this study on 25 brain antigen-directed AB measured in serum was that the location of the epitope (intracellular versus extracellular) obviously determines the Ig class, with intracellular epitopes predisposing to IgG. This may at least question the widely believed exclusive association of IgG with pathology [18].

HIGH SEROPREVALENCE OF *N*-METHYL-D-ASPARTATE RECEPTOR 1-AB ACROSS HEALTH AND DISEASE

An interesting example in this regard and therefore in the focus of the present article are AB against the *N*-methyl-*d*-aspartate receptor 1 (NMDAR1-AB; *new nomenclature, GluN1, disregarded here for consistency with AB literature*). NMDAR exist as di-heteromeric or tri-heteromeric complexes, with the composition of the four subunits determining receptor properties [19–21]. As NMDAR1 is an obligatory partner in all possible constellations, AB against this subunit may potentially affect all NMDAR in brain and periphery.

NMDAR1-AB of the IgG class were originally reported in a condition called 'anti-NMDAR encephalitis' and claimed to be disease-pathognomonic [22–24]. The array of symptoms described in this condition is somewhat variable but essentially consistent with NMDAR1 antagonism as inducible by for example the NMDAR antagonist ketamine: Psychosis, epileptic seizures, extrapyramidal movement disorders, cognitive decline, reduced consciousness and autonomic dysfunction. In none of the earlier reports on 'anti-NMDAR encephalitis', healthy controls were analyzed to support

the claim of disease-specificity. Therefore, the age-dependent seroprevalence of up to >20% NMDAR1-AB in healthy subjects as well as across all investigated disease groups (neuropsychiatric as well as other medical conditions like hypertension and diabetes) came as surprise [25–28]. Compared with other brain-antigen directed AB (mostly <2%), NMDAR1-AB have the highest disease-independent seroprevalence [18]. Serum titers as well as the distribution of immunoglobulin classes (mainly IgM and IgA; IgG rarest; IgE not detected so far) are similar in health and any to date investigated disease [27–29,30▪▪]. Many reports, including recent ones, describing either higher seroprevalence in certain disorders and/or absence/scarcity in a particular disease/health group are based on much too small numbers of subjects for a firm conclusion to be drawn (e.g. [31▪,32▪]). Among them are case reports, often without mentioning immunoglobulin classes, titers, or whether serum or cerebrospinal fluid (CSF) was analyzed. Inadequate or uncritical citation of the literature is prevailing [33▪,34▪].

In a heterogeneous group of 323 cancer patients, the seroprevalence of neuronal surface AB (IgA, IgM) was reported to be high [32▪], but is actually in the expected range of the investigated age (NMDAR1-AB 16.7%; mean age 62 years) [18,27,28]. The number of controls in this study ($N = 65$ neurological disease controls and $N = 40$ healthy blood donors) is too low for solid comparative conclusions. Cognitive deficits described by this study in AB-positive patients with chronic BBB dysfunction (pathological albumin quotient) are an interesting finding, even though not unexpected (chronic ketamine-like effects), but unlikely due to specific features of any particular cancer [32▪]. On the other hand, some cancers may well lead – via as yet widely unknown mediators – to a persistently perturbed BBB and thus allow greater access of circulating AB to the brain.

FUNCTIONALITY AND EPITOPES OF *N*-METHYL-D-ASPARTATE RECEPTOR 1-AB IN HEALTH AND DISEASE

All NMDAR1-AB, independent of the immunoglobulin class, proved to be functional in human and mouse, both *in vivo* [27,28] and *in vitro* [27,30▪▪,35], next raising the question of target epitopes. Published work on NMDAR1-AB epitopes had been limited to IgG recognizing NTD and NTD-G7 domain (N368/G369), originally deemed pathognomonic for 'anti-NMDAR encephalitis' [22,36]. Thus, for the first time, NMDAR1-AB of three immunoglobulin classes (IgM, IgG, IgA), derived from randomly selected individuals (out of thousands) of different age, sex and medical condition, were tested regarding in-vitro functionality and epitope location [30▪▪]. All NMDAR1-AB positive sera led to NMDAR1 internalization in inducible pluripotent stem cell (IPSC)-derived human cortical neurons and to reduced glutamate-evoked response in NR1–1b/NR2A-expressing oocytes. Several different epitopes recognized by NMDAR1-AB (all classes) were identified, located in the extracellular part of NMDAR1 (NTD, LBD) or intracellular, C-terminal (CTD) and in the extralarge pore domain (xlp). No consistent functional or epitope pattern was detectable regarding immunoglobulin class or health/disease state. Half of the screened sera showed polyclonality/polyspecificity, the other half was monoclonal or oligoclonal/oligospecific (mainly IgG) [30▪▪]. Factors predisposing young women to neuropsychiatric manifestations of NMDAR-associated autoimmunity, for example in lupus erythematosus [37] or anti-NMDAR encephalitis [22,36], may indeed be related to NTD or NTD-G7 epitopes.

Different functionality of NMDAR1-AB (IgG) derived from schizophrenic as compared with healthy subjects has been reported by Jezequel *et al.* [38▪▪]. Super-resolution imaging was used to provide a nanoscale surface organization map of NMDAR in hippocampal networks. The authors describe specific alterations of the NMDAR synaptic trafficking by NMDAR1-AB from four psychotic patients. According to their hypothesis, NMDAR hypofunction in schizophrenia might be induced by destabilizing synaptic NMDAR and its interacting partner EphB2R. In contrast, in their control conditions (1 AB− individual; 3 healthy AB+ subjects), NMDAR laterally diffuse and stabilize in nanometer-sized clusters within glutamatergic synapses [38▪▪]. Another article of these authors, using the same method, finds one NMDAR1-AB carrying autistic individual not to differ from one healthy carrier [39▪]. Even though highly interesting and based on a sophisticated method, more work is needed to confirm any psychosis-relation of these findings. NMDAR1-AB were not characterized by epitope mapping and questions of oligospecificity or polyspecificity or presence of other immunoglobulin classes (apart from IgG) remained open [38▪▪]. What, if not the target epitope of AB, would explain the influence on synaptic anchoring in a standardized experiment?

Considering the low seroprevalence of only ~1% IgG NMDAR1-AB based on more than 5000 individuals investigated across health and disease [40], the authors were incredibly lucky to find any IgG+ subject when screening only 48 schizophrenic [38▪▪] or 24 autistic patients and 18 healthy controls

[39▪] for the presence of NMDAR-AB. Even when testing 104 healthy controls, three IgG+ individuals are unusual [38▪▪]. In contrast, a recent report on 78 and 234 schizophrenic subjects, respectively, did not find a single IgG+ patient [31▪]. These by chance findings underline the above mentioned problems of seroprevalence estimates based on too small numbers.

'ANTI-NMDAR ENCEPHALITIS' IS UNLIKELY A SEPARATE DISEASE ENTITY

Several current articles failed to confirm earlier 'exclusivity claims' of 'anti-NMDAR encephalitis'. Extreme delta brush had been interpreted as unique EEG pattern in adults with 'anti-NMDAR encephalitis' [41], a statement now invalidated [42▪]. Addressing the predisposition of AB carriers to epilepsy, high neurological AB prevalence (34.8% neurological AB; 3.6% NMDAR1-AB; pretreatment sera) was described in prospectively evaluated cases with epilepsy of unknown cause [43▪▪]. AB+ patients showed greater likelihood of neuropsychiatric symptoms or autonomic dysfunction. Also in this study, immunoglobulin class and method of AB detection were not mentioned, BBB function not discussed, and the high overall AB seroprevalence in health and disease not considered.

Another recent claim is the 'characteristic pattern of whole-brain functional connectivity alterations in anti-NMDAR encephalitis that is well suited to explain the major clinical symptoms of the disorder' [44▪▪]. The authors investigated at highly variable time points after the initial diagnosis 43 'anti-NMDAR encephalitis' cases by resting state fMRI. A large proportion of subjects had at the day of fMRI negative NMDAR1-AB titers [44▪▪]. Thus, any conclusion linking the described dysconnectivity to NMDAR1-AB is problematic. Long-term persisting effects of NMDAR1-AB once they are eliminated by e.g. immunosuppression or plasmapheresis, have not yet been documented anywhere. Experimental approaches rather point to a rapid loss of effects after AB vanishing [45]. Even more importantly, the study lacks a well matched group of encephalitis patients without history of NMDAR1-AB as adequate control. A connectivity disturbance is foreseeable for any kind of encephalitis [46▪▪].

Along these lines and enlightening for clinicians, no appreciable differences were extracted by comparing NMDAR1-AB+ and NMDAR1-AB− encephalitis cases, except for typical NMDAR antagonistic (ketamine-like) symptoms. CSF analysis, EEG monitoring, MRI scanning, accompanying tumor frequency and outcomes were not different [47]. These findings would support that 'anti-NMDA encephalitis' is not a separate condition, but marks any type of encephalitis in which the highly prevalent NMDAR1-AB happen to be present, are boosted, and dramatically shape the clinical picture. Dependent on the circumstances, namely absence or presence of an underlying brain inflammation, NMDAR1-AB may contribute to chronic processes (dementia, psychosis, epilepsy of unknown cause, personality changes) or fulminant courses ('anti-NMDAR encephalitis'), provided access to the brain (BBB dysfunction or intrathecal synthesis). The prodromal phase of many encephalitides is 'infectious-like' [22–24,47], and infections cannot easily be excluded by routine analyses. Later symptoms may be 'ketamine-like' due to the presence of NMDAR1-AB among many other brain antigen-directed AB [45,48]. The underlying condition finally determines the highly variable clinical and pathological picture, namely further symptoms, diagnostic readouts (EEG, MRI, CSF/blood parameters), treatment response, outcome and autopsy findings. It may be problematic if the search for causes stops after detection of NMDAR1-AB (IgG). We note, however, that in most cases, 'polypragmatic' treatment anyhow includes antibiotics, antiviral compounds and corticosteroids/immunosuppressants on top of a wide range of supporting measures.

The question whether NMDAR1-AB can induce inflammation in the brain has recently been experimentally addressed [49▪▪]. Active immunization of mice against four peptides of the extracellular NMDAR1 domain (including NTD-G7; N368/G369) leads to high circulating levels of specific AB. After 4 weeks, the endogenously formed NMDAR1-AB (IgG) induce psychosis-like symptoms upon MK-801 challenge in $ApoE^{-/-}$ mice, characterized by open BBB, but not in $ApoE^{+/+}$ littermates, which are indistinguishable from ovalbumin controls. Most importantly, NMDAR1-AB do not induce any sign of inflammation in the brain. Immunohistochemical staining for microglial activation markers and T-lymphocytes yields comparable results, irrespective of immunization. Thus, NMDAR1-AB (here IgG) shape behavioral phenotypes upon access to the brain but do not cause encephalitis [49▪▪]. This result may put recent reports into perspective that analyzed plasma cell clones from CSF of encephalitis patients and purport that 'human CSF monoclonal NMDAR1-AB are sufficient for encephalitis pathogenesis' [48] or claim to have proven their pathogenic relevance by passive transfer of disease symptoms from man to mouse, based on a single behavior test, novel object recognition, but no histology [45].

INCREASING SEROPREVALENCE OF *N*-METHYL-D-ASPARTATE RECEPTOR 1-AB WITH AGE: WHAT MAY BE THE (PATHO) PHYSIOLOGICAL ROLE AND WHICH ARE THE INDUCERS?

The increasing seroprevalence of NMDAR1-AB with age is particularly interesting, as it is in contrast to most studies in humans and rodents that report natural AB of the IgM class to decrease or lose their effectiveness with age (reviewed in [1]). At least for NMDAR1-AB, this is clearly not the case, as all classes (IgM, IgA, IgG) rise with age [27,28,49▪▪]. The reason for this discrepancy is still obscure, but induction of NMDAR1-AB by for example influenza infection [27,28] or by chronic life stress [49▪▪], stimulants with accumulating relevance upon aging, may partly explain this finding.

Remarkably, not only humans, but all other mammals investigated, dogs, cats, mice, rats and monkeys, show high seroprevalence of NMDAR1-AB, with Ig class distribution comparable with humans [49▪▪]. We note that NMDAR1 has >99% sequence homology across mammals. Functionality of NMDAR1-AB was proven for all species by NMDAR endocytosis in human IPSC-derived cortical neurons. However, the age dependence was lost in monkeys (rhesus macaques and baboons) in which already young animals, captured/transported or born in captivity, displayed high seroprevalence. Hypothesizing that this finding may be related to chronic life stress, for example caused by an unfamiliar or unfriendly environment, we screened human migrants and likewise found that the age-dependence had disappeared. In both migrants and monkeys, IgA was the predominant class accounting for the early AB rise [49▪▪]. Mechanisms behind the selective increase in IgA still need clarification. Nevertheless, it fits well to early work showing distinct serum IgA response to stress [50].

In the context of natural as well as pathological NMDAR1-AB, the emerging role of NMDAR in the immune system itself is intriguing: NMDAR antagonists dampen B cell [51] as well as T-cell functions [52]. NMDAR is rapidly upregulated upon CD4+ T-cell activation in humans and neurotransmitter/agonist signaling via NMDAR leads to decreased T helper type 1-like and enhanced T helper type 2-like immune balance, affecting proliferation, cytokine production and cell survival [53▪▪]. Even though ultimate proof of NMDAR1 functionality on blood cells is still pending, it is intriguing to speculate that NMDAR1-AB as humoral (auto)immunity players could feedback-modulate the immune system.

Many other questions in the field of humoral autoimmunity in general and of NMDAR1-AB in particular remain open: The old question [5], raised again in a recent review [54], of how brain constituents get antigens, is pivotal also for NMDAR1-AB. Central nervous system constituents are naturally not antigenic but may belong to the normal autoimmune repertoire of mammals and/or be converted to be immunogens via release of brain debris into circulation, formation of complexes with other substances or alteration of configuration [55], posttranslational protein modification, for example citrullination [56,57], or exposure in 'free solution form' to AB as a result of cellular injury or death [54,58]. These and other mechanisms, for example molecular mimicry, may account for the predisposition observed upon oncological conditions (teratoma; [22]), infections (e.g. influenza A and B), genetic constellation (SNP-rs524991, with an adjacent gene being NMDAR biology-related) or chronic life stress [27,28,49▪▪]. Exposure of the brain to the immune system via leaky BBB alone, however, may not suffice to induce NMDAR1-AB formation, as *APOE4* carrier status or *ApoE* knockout alone, does not seem to predispose to NMDAR1-AB formation [28,35,49▪▪].

CONCLUSION

All naturally occurring NMDAR1-AB, irrespective of immunoglobulin isotype or epitope, are potentially syndrome-relevant upon access to or production within the brain. The intriguing fact that NMDAR1-AB age-dependently increase to an extremely high seroprevalence across mammals may indicate additional not yet understood roles in natural/physiological autoimmunity.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:
- ▪ of special interest
- ▪▪ of outstanding interest

1. Lobo PI. Role of natural autoantibodies and natural IgM anti-leucocyte autoantibodies in health and disease. Front Immunol 2016; 7:198.

2. Nguyen TT, Baumgarth N. Natural IgM and the development of B cell-mediated autoimmune diseases. Crit Rev Immunol 2016; 36:163–177.
3. Wienands J, Engels N. Control of memory B cell responses by extrinsic and intrinsic mechanisms. Immunol Lett 2016; 178:27–30.
4. Wienands J, Engels N. The memory function of the B cell antigen receptor. Curr Top Microbiol Immunol 2016; 393:107–121.
5. Ingram CR, Phegan KJ, Blumenthal HT. Significance of an aging-linked neuron binding gamma globulin fraction of human sera. J Gerontol 1974; 29:20–27.
6. Nagele RG, Clifford PM, Siu G, *et al.* Brain-reactive autoantibodies prevalent in human sera increase intraneuronal amyloid-β(1–42) deposition. J Alzheimers Dis 2011; 25:605–622.
7. Han M, Nagele E, DeMarshall C, *et al.* Diagnosis of Parkinson's disease based on disease-specific autoantibody profiles in human sera. PLoS One 2012; 7:e32383.
8. ■ Kannan G, Gressitt KL, Yang S, *et al.* Pathogen-mediated NMDA receptor autoimmunity and cellular barrier dysfunction in schizophrenia. Transl Psychiatry 2017; 7:e1186.

Hint by ELISA that exposure to the pathogen *T. gondii* in mouse and human produces an anti-NMDAR2 immune response accompanied by endothelial barrier dysfunction in both gut and CNS.

9. Nagele E, Han M, Demarshall C, *et al.* Diagnosis of Alzheimer's disease based on disease-specific autoantibody profiles in human sera. PLoS One 2011; 6:e23112.
10. Putterman C, Wu A, Reiner-Benaim A, *et al.* SLE-key® rule-out serologic test for excluding the diagnosis of systemic lupus erythematosus: developing the ImmunArray iCHIP®. J Immunol Methods 2016; 429:1–6.
11. ■ Balint B, Vincent A, Meinck HM, *et al.* Movement disorders with neuronal antibodies: syndromic approach, genetic parallels and pathophysiology. Brain 2018; 141:13–36.

Up-to-date review for the interested reader.

12. Coutinho E, Harrison P, Vincent A. Do neuronal autoantibodies cause psychosis? A neuroimmunological perspective. Biol Psychiatry 2014; 75:269–275.
13. Crisp SJ, Kullmann DM, Vincent A. Autoimmune synaptopathies. Nat Rev Neurosci 2016; 17:103–117.
14. Diamond B, Huerta PT, Mina-Osorio P, *et al.* Losing your nerves? Maybe it's the antibodies. Nat Rev Immunol 2009; 9:449–456.
15. ■ Fukata M, Yokoi N, Fukata Y. Neurobiology of autoimmune encephalitis. Curr Opin Neurobiol 2017; 48:1–8.

Recent review for the interested reader; however, coverage of important aspects and critical evaluation lacking.

16. Höftberger R, Lassmann H. Chapter 20 – immune-mediated disorders. In: Kovacs GG, Alafuzoff I, editors. Handbook of clinical neurology. Amsterdam, Netherlands: Elsevier; 2017. pp. 285–299.
17. ■ Mader S, Brimberg L, Diamond B. The role of brain-reactive autoantibodies in brain pathology and cognitive impairment. Front Immunol 2017; 8:1101.

Up-to-date review for the interested reader.

18. Dahm L, Ott C, Steiner J, *et al.* Seroprevalence of autoantibodies against brain antigens in health and disease. Ann Neurol 2014; 76:82–94.
19. Hansen KB, Yi F, Perszyk RE, *et al.* NMDA receptors in the central nervous system. Methods Mol Biol 2017; 1677:1–80.
20. Lau CG, Zukin RS. NMDA receptor trafficking in synaptic plasticity and neuropsychiatric disorders. Nat Rev Neurosci 2007; 8:413–426.
21. Li F, Tsien JZ. Memory and the NMDA receptors. N Engl J Med 2009; 361:302–303.
22. Dalmau J, Gleichman AJ, Hughes EG, *et al.* Anti-NMDA-receptor encephalitis: case series and analysis of the effects of antibodies. Lancet Neurol 2008; 7:1091–1098.
23. Dalmau J, Lancaster E, Martinez-Hernandez E, *et al.* Clinical experience and laboratory investigations in patients with anti-NMDAR encephalitis. Lancet Neurol 2011; 10:63–74.
24. Titulaer MJ, McCracken L, Gabilondo I, *et al.* Treatment and prognostic factors for long-term outcome in patients with anti-NMDA receptor encephalitis: an observational cohort study. Lancet Neurol 2013; 12: 157–165.
25. Steiner J, Walter M, Glanz W, *et al.* Increased prevalence of diverse *N*-methyl-D-aspartate glutamate receptor antibodies in patients with an initial diagnosis of schizophrenia: specific relevance of IgG NR1a antibodies for distinction from *N*-methyl-D-aspartate glutamate receptor encephalitis. JAMA Psychiatry 2013; 70:271–278.
26. Steiner J, Teegen B, Schiltz K, *et al.* Prevalence of *N*-methyl-D-aspartate receptor autoantibodies in the peripheral blood: healthy control samples revisited. JAMA Psychiatry 2014; 71:838–839.
27. Hammer C, Stepniak B, Schneider A, *et al.* Neuropsychiatric disease relevance of circulating anti-NMDA receptor autoantibodies depends on blood–brain barrier integrity. Mol Psychiatry 2014; 19:1143–1149.
28. Zerche M, Weissenborn K, Ott C, *et al.* Preexisting serum autoantibodies against the NMDAR subunit NR1 modulate evolution of lesion size in acute ischemic stroke. Stroke 2015; 46:1180–1186.
29. Castillo-Gomez E, Kastner A, Steiner J, *et al.* The brain as immunoprecipitator of serum autoantibodies against *N*-methyl-D-aspartate receptor subunit NR1. Ann Neurol 2016; 79:144–151.
30. ■■ Castillo-Gomez E, Oliveira B, Tapken D, *et al.* All naturally occurring autoantibodies against the NMDA receptor subunit NR1 have pathogenic potential irrespective of epitope and immunoglobulin class. Mol Psychiatry 2017; 22:1776–1784.

First systematic investigation of epitopes in parallel with functionality (internalization assay and electrophysiology) of NMDAR1-AB of all classes from ill as well as healthy subjects.

31. ■ Chen CH, Cheng MC, Liu CM, *et al.* Seroprevalence survey of selective antineuronal autoantibodies in patients with first-episode schizophrenia and chronic schizophrenia. Schizophr Res 2017; 190:28–31.

Article on small numbers of patients with schizophrenia that does not detect NMDAR1-AB of the IgG class. No real surprise (statistics of small numbers). Other classes not determined nor discussed.

32. ■ Finke C, Bartels F, Lutt A, *et al.* High prevalence of neuronal surface autoantibodies associated with cognitive deficits in cancer patients. J Neurol 2017; 264:1968–1977.

Article on patients with different cancers and small control groups. Cognitive decline in AB+ patients with BBB dysfunction is interesting but unlikely cancer-specific.

33. ■ Escudero D, Guasp M, Arino H, *et al.* Antibody-associated CNS syndromes without signs of inflammation in the elderly. Neurology 2017; 89:1471–1475.

Article on 35 aged subjects with neurologic syndromes and AB against neuronal surface antigens. Unclear whether AB were measured in serum or CSF, titers or immunoglobulin classes not mentioned; no controls; role of AB in these conditions remains unclear.

34. ■ Zhou L, ZhangBao J, Li H, *et al.* Cerebral cortical encephalitis followed by recurrent CNS demyelination in a patient with concomitant anti-MOG and anti-NMDA receptor antibodies. Mult Scler Relat Disord 2017; 18:90–92.

A sample case report out of many published every year. NMDAR1-AB induced too late to explain any causal connection with encephalitis. Immunoglobulin class not mentioned. Titer unclear.

35. Hammer C, Zerche M, Schneider A, *et al.* Apolipoprotein E4 carrier status plus circulating anti-NMDAR1 autoantibodies: association with schizoaffective disorder. Mol Psychiatry 2014; 19:1054–1056.
36. Gleichman AJ, Spruce LA, Dalmau J, *et al.* Anti-NMDA receptor encephalitis antibody binding is dependent on amino acid identity of a small region within the GluN1 amino terminal domain. J Neurosci 2012; 32: 11082–11094.
37. Ogawa E, Nagai T, Sakuma Y, *et al.* Association of antibodies to the NR1 subunit of *N*-methyl-D-aspartate receptors with neuropsychiatric systemic lupus erythematosus. Mod Rheumatol 2016; 26:377–383.
38. ■■ Jezequel J, Johansson EM, Dupuis JP, *et al.* Dynamic disorganization of synaptic NMDA receptors triggered by autoantibodies from psychotic patients. Nat Commun 2017; 8:1791.

A sophisticated novel super-resolution imaging approach allowed the detection of alterations in NMDAR synaptic trafficking by NMDAR1-AB (IgG) from four psychotic patients but not from three controls. Epitopes to help explain these findings not yet determined.

39. ■ Grea H, Scheid I, Gaman A, *et al.* Clinical and autoimmune features of a patient with autism spectrum disorder seropositive for anti-NMDA-receptor autoantibody. Dialogues Clin Neurosci 2017; 19:65–70.

Another article on the imaging method used in [38■■], including one control and one autistic individual.

40. Ehrenreich H. Autoantibodies against the *N*-methyl-*d*-aspartate receptor subunit NR1: untangling apparent inconsistencies for clinical practice. Front Immunol 2017; 8:181.
41. Schmitt SE, Pargeon K, Frechette ES, *et al.* Extreme delta brush: a unique EEG pattern in adults with anti-NMDA receptor encephalitis. Neurology 2012; 79:1094–1100.
42. ■ Baykan B, Gungor Tuncer O, Vanli-Yavuz EN, *et al.* Delta brush pattern is not unique to NMDAR encephalitis: evaluation of two independent long-term EEG cohorts. Clin EEG Neurosci 2017. [Epub ahead of print]

Article on EEG, demonstrating delta brush pattern in intensive care unit patients with multiple different diagnoses.

43. ■■ Dubey D, Alqallaf A, Hays R, *et al.* Neurological autoantibody prevalence in epilepsy of unknown etiology. JAMA Neurol 2017; 74:397–402.

Prospective study reporting in pretreatment sera of 112 patients with epilepsy of unknown cause 34.8% neurological AB (3.6% NMDAR1-AB); AB+ patients show clinical correlates; immunoglobulin class and methods of determination not reported.

44. ■■ Peer M, Pruss H, Ben-Dayan I, *et al.* Functional connectivity of large-scale brain networks in patients with anti-NMDA receptor encephalitis: an observational study. Lancet Psychiatry 2017; 4:768–774.

Connectivity alterations seen by resting state fMRI in 'anti-NMDAR encephalitis' are believed to explain clinical symptoms; however, evidence of persisting effects of AB after their elimination and of specific association with 'anti-NMDAR encephalitis' still missing.

45. Malviya M, Barman S, Golombeck KS, *et al.* NMDAR encephalitis: passive transfer from man to mouse by a recombinant antibody. Ann Clin Transl Neurol 2017; 4:768–783.
46. ■■ Oliveira B, Ehrenreich H. Pursuing functional connectivity in NMDAR1 autoantibody carriers. Lancet Psychiatry 2018; 5:21–22.

Letter that makes readers aware of the lack of adequate controls in [45].

47. Chen X, Li JM, Liu F, *et al.* Anti-*N*-methyl-D-aspartate receptor encephalitis: a common cause of encephalitis in the intensive care unit. Neurol Sci 2016; 37:1993–1998.

48. Kreye J, Wenke NK, Chayka M, *et al.* Human cerebrospinal fluid monoclonal *N*-methyl-D-aspartate receptor autoantibodies are sufficient for encephalitis pathogenesis. Brain 2016; 139:2641–2652.

49. ■■ Pan H, Oliveira B, Saher G, *et al.* Uncoupling the widespread occurrence of anti-NMDAR1 autoantibodies from neuropsychiatric disease in a novel autoimmune model. Mol Psychiatry 2018.

Article reporting high abundancy of NMDAR1-AB of all immunoglobulin classes in mammals and potential induction by chronic life stress; immunization of mice with open BBB, producing highest circulating NMDAR1-AB (IgG) titers, does not cause brain inflammation.

50. Maes M, Hendriks D, Van Gastel A, *et al.* Effects of psychological stress on serum immunoglobulin, complement and acute phase protein concentrations in normal volunteers. Psychoneuroendocrinology 1997; 22:397–409.

51. Simma N, Bose T, Kahlfuss S, *et al.* NMDA-receptor antagonists block B-cell function but foster IL-10 production in BCR/CD40-activated B cells. Cell Commun Signal 2014; 12:75.

52. Lowinus T, Bose T, Busse S, *et al.* Immunomodulation by memantine in therapy of Alzheimer's disease is mediated through inhibition of Kv1.3 channels and T cell responsiveness. Oncotarget 2016; 7:53797–53807.

53. ■■ Orihara K, Odemuyiwa SO, Stefura WP, *et al.* Neurotransmitter signalling via NMDA receptors leads to decreased T helper type 1-like and enhanced T helper type 2-like immune balance in humans. Immunology 2017. [Epub ahead of print]

Article on expression and potential function of NMDAR in immune cells: NMDAR is apparently rapidly upregulated upon CD4+ T-cell activation, and agonist signaling via NMDAR leads to decreased Th-1-like and enhanced Th-2-like immune balance.

54. Kobeissy F, Moshourab RA. Autoantibodies in CNS trauma and neuropsychiatric disorders: a new generation of biomarkers. In: Kobeissy FH, editor. Brain neurotrauma: molecular, neuropsychological, and rehabilitation aspects. Boca Raton (FC): CRC Press/Taylor & Francis; 2015.

55. Behan PO, Lowenstein LM, Stilmant M, *et al.* Landry–Guillain–Barre–Strohl syndrome and immune-complex nephritis. Lancet 1973; 1: 850–854.

56. Acharya NK, Nagele EP, Han M, *et al.* Neuronal PAD4 expression and protein citrullination: possible role in production of autoantibodies associated with neurodegenerative disease. J Autoimmun 2012; 38:369–380.

57. Acharya NK, Nagele EP, Han M, *et al.* Autoantibodies: double agents in human disease. Sci Transl Med 2013; 5:186fs119.

58. Ankeny DP, Popovich PG. B cells and autoantibodies: complex roles in CNS injury. Trends Immunol 2010; 31:332–338.