2015

Integrative neuroscience approach to neuropsychiatric lupus

P. T. Huerta
Zucker School of Medicine at Hofstra/Northwell

E. L. Gibson
Northwell Health

C. Rey
Northwell Health

T. S. Huerta
Northwell Health

HHS Public Access

Author manuscript

Immunol Res. Author manuscript; available in PMC 2016 June 23.

Published in final edited form as:

Immunol Res. 2015 December ; 63(1-3): 11–17. doi:10.1007/s12026-015-8713-6.

Integrative neuroscience approach to neuropsychiatric lupus

Patricio T. Huerta[1,2], **Elizabeth L. Gibson**[1], **Carson Rey**[1], **Tomás S. Huerta**[1], and **Patricio T. Huerta**

Patricio T. Huerta: pato.huerta@gmail.com

[1]Laboratory of Immune and Neural Networks, Feinstein Institute for Medical Research, 350 Community Drive, Manhasset, NY 11030, USA

[2]Department of Molecular Medicine, Hofstra North Shore LIJ Medical School, Hempstead, NY, USA

Abstract

We present a succinct review of our approach to study the interactions between the DNA-reactive antibodies that cross-react with the GluN2A and GluN2B subunits of the *N*-methyl-D-aspartate receptor, denoted DNRABs, and their brain targets in subjects with neuropsychiatric systemic lupus erythematosus (NPSLE). We have analyzed the DNRAB-based brain symptomatology in mouse models of NPSLE by using an integrative neuroscience approach, which includes behavioral assessment coupled with electrophysiological studies of neural networks and synaptic connections in target brain regions, such as the CA1 region of the hippocampus. Our results suggest a framework for understanding the interactions between immune factors and neural networks.

Keywords

NMDAR; Mouse; Hippocampus; Cognition; Brain autoimmunity

NPSLE and cognitive impairment

Systemic lupus erythematosus (SLE) is a chronic immune disorder that affects many organs in the body. At least 5 million individuals suffer SLE worldwide, with up to 90 % of them being women of childbearing age. In the USA, it occurs at higher frequency in populations of African American, Hispanic, and Asian American origin [1]. Neuropsychiatric systemic lupus erythematosus (NPSLE) refers to the collection of syndromes, ranging from psychosis to memory problems, which target the nervous system in 40–90 % of subjects with SLE [2–9]. Many NPSLE manifestations occur at the onset or within the first year of SLE diagnosis, and are uncorrelated with active systemic disease and serologic activity [10]. NPSLE is accompanied by a variable burden of disability and significantly diminishes quality of life.

Correspondence to: Patricio T. Huerta, pato.huerta@gmail.com.

Patricio T. Huerta, Elizabeth L. Gibson, Carson Rey and Tomás S. Huerta have contributed equally to this work.

NPSLE can be identified with the use of 19 criteria established by an expert committee of the American College of Rheumatology (ACR); twelve of which affect the central nervous system (CNS) and seven the peripheral nervous system [3]. CNS involvement can be diffuse (as in the cases of cognitive dysfunction, mood disorder, anxiety disorder, and psychosis), focal (as in cerebrovascular disease), or complex (mixture of diffuse and focal). However, the bewildering array of reversible and irreversible symptoms attributable to brain dysfunction and caused by NPSLE has hampered a mechanistic understanding. It is therefore critical to explore innovative paradigms to understand the pathophysiological mechanisms of this vexing problem in SLE.

Cognitive impairment (CI) is highly prevalent in NPSLE, ranging from 20 to 80 % [4–7, 11–17]. Loss of cognitive function, as defined by the ACR, is characterized by impairment in one or more of the following cognitive domains: simple attention, complex attention, memory, visual-spatial processing, language, reasoning, problem solving, psychomotor speed, and executive functions. Most studies suggest a slow cognitive decline and no association with disease activity [18–21]. It is somewhat perplexing that the wide-ranging prevalence of CI reflects the methodological variations by which it has been measured rather than biological differences [17, 22–24]. The course of CI has been described as evanescent and fluctuating over a 2- to 5-year period [25, 26] but progressing slowly over longer periods, and even reaching the severity of dementia in 3–5 % of subjects with SLE [25–29]. Unambiguous CI signs can be challenging to ascertain clinically [29], but strict adherence to the ACR standard (which proposes a 1-h battery of neuropsychological tests) together with the emergence of improved cognitive tests might yield more exact CI estimates in NPSLE patients.

Brain-reactive antibodies in NPSLE

The pathogenesis of NPSLE has been linked with several immune factors, such as inflammatory cytokines, autoantibodies, and immune complexes. For instance, cerebrovascular disease in NPSLE, a focal type of insult, may result from vascular insults (such as thrombosis) mediated by anti-phospholipid antibodies, accelerated atherosclerosis, and the non-deformability of red blood cells that are coated with immune complexes [30–32].

Early studies have demonstrated that serum antibodies from subjects with SLE bind to normal brain tissue [33–38]. The occurrence of antibodies capable of altering the function of neuronal cells offers a promising avenue to study the diffuse CNS syndromes associated with NPSLE. Indeed, an increasing number of studies have strengthened the idea that some non-focal symptoms can be linked to the presence of DNRABs, a subclass of DNA-reactive antibodies that bind the GluN2A and GluN2B subunits of the *n*-methyl-d-as-partate receptor (NMDAR) with very high specificity and do not appear to bind any other brain molecule [39–49]. There is now compelling evidence for the pathogenic role of DNRABs in NPSLE [50–55] suggesting that these antibodies can cause more than one manifestation, specifically memory dysfunction and emotional disturbance, depending on the mechanisms by which DNRABs penetrate the blood–brain barrier (BBB) that separates the brain from circulation. Several investigators have started using these paradigms to test brain effects of other SLE-

related autoantibodies [56–59]. Interestingly, these antibody molecules are likely to be polyreactive, that is, capable of binding a variety of structurally unrelated brain antigens.

NMDAR: synaptic receptor targeted by NPSLE

We have developed a strategy to study the effects of DNRABs in the brain based on the seminal finding that these autoantibodies recognize a linear 5-amino acid sequence consisting of D/E, W, D/E, Y, and S/G [39]. This epitope is localized in the extracellular, amino-terminal domains of GluN2A (residues 283–287, sequence DWDYS) and GluN2B (residues 284–288, sequence EWDYG); each subunit is an essential component of the NMDAR [60].

NMDARs are localized within the most abundant synapses of the forebrain, which use glutamate as the neurotransmitter, and connect excitatory neurons in brain regions such as the cerebral cortex, the hippocampus, and the amygdala [61–63]. During excitatory synaptic transmission, glutamate is released from presynaptic terminals and binds to ligand-gated receptors on the postsynaptic membrane, the most common of which are the NMDARs [61–63] and the amino-3-hydroxy-5-methylisoxazole-4-propionic acid receptors (AMPARs) [64]. The latter mediate fast excitation, through the influx of sodium into the postsynaptic cell, which results in rapid depolarization of the postsynaptic membrane. NMDARs mediate a slower and longer-lasting excitation through the influx of calcium. Normally, NMDARs participate in synaptic plasticity largely because of their high calcium permeability and slow deactivation and desensitization kinetics [61–63, 65–68]. These very same properties, however, allow NMDARs to trigger excitotoxicity and apoptosis under abnormal conditions [69–71].

In the forebrain, functional NMDARs are assembled as a pair of GluN1 subunits and a pair of GluN2 subunits. GluN1s are essential channel-forming subunits, and they bind glycine and determine calcium permeability [65], while GluN2 subunits bind glutamate and contain regions that regulate deactivation and desensitization [65]. NMDARs are also voltage-dependent, becoming activated only during significant postsynaptic depolarization. In the resting state, the pore of the ion channel formed by the NMDAR is blocked by magnesium, which during the depolarized state is forced out of the pore, allowing calcium to enter into the postsynaptic cell [68]. NMDARs can be thought as associative gates, requiring presynaptic activity (glutamate) and postsynaptic activity (depolarization) to be activated [68]. Moreover, electrophysiological studies have determined that the response of GluN1–GluN2A complexes is short (~100 ms) when compared to GluN1–GluN2B complexes (~250 ms), implying that the latter lead to higher calcium influx into the postsynaptic cell [72].

NMDARs in the hippocampus are essential for the acquisition of certain types of memory. The hippocampus is organized into interconnected regions: the dentate gyrus (DG), the cornu ammonis (CA) areas (CA1, CA2, and CA3), and the subiculum [73, 74]. These areas receive projections from, and project back to, the cerebral cortex. Much research has focused on the hippocampus to provide insight into the neural substrates of memory. It is well documented that humans with damage to this brain region exhibit episodic memory impairment, but can otherwise function normally [74–76]. Similarly, rodents with

hippocampal lesions are severely impaired in tasks that depend on memories in the spatial and temporal domains [78–82]. With the use of genetic techniques, we have demonstrated that mice in which the NMDARs have been deleted from the CA1 pyramidal cells are unable to establish spatial and temporal memories [81, 82], confirming the lesion studies and providing more detailed information on the molecular requirements for hippocampal memory function.

Integrative neuroscience applied to NPSLE

We have analyzed the effects of DNRABs at different levels of neural organization, from molecules and synapses to brain regions and whole-brain (Fig. 1), and have identified alterations that, in combination, provide a multilevel mechanism for the deleterious action of DNRABs within the brain. The pioneering work by Betty Diamond and collaborators [39] identified cross-reactive antigens for anti-double-stranded deoxyribonucleic acid (anti-dsDNA) antibodies; they found a nephritogenic mouse monoclonal anti-dsDNA antibody, termed R4A, which was reacted with a phage peptide display library harboring random 10 amino acid inserts encoded by random 30-base pair insets. A consensus sequence for R4A (D/E-W-D/E-Y-S/G) was identified in 36 phage clones. Of these, 23 phage clones had identical DWEYSVWLSN amino acid sequences. A protein database search revealed that the DWEYS consensus sequence was found in some bacterial antigens but more surprisingly, also present in the N-terminal domains of mouse, rat, and human GluN2A and GluN2B, but not in GluN2C and GluN2D [39].

We have developed an in vivo model in which BALB/c mice synthesize DNRAbs following immunization with a configuration of the DWEYS sequence multimerized on a polylysine backbone (termed MAP-DWEYS), while BALB/c mice immunized with the polylysine backbone alone (MAP-core) do not [40]. This model allows us to evaluate DNRAbs as causal agents of neuronal injury, independent of other autoantibodies, and the high levels of systemic inflammatory mediators found in spontaneous mouse SLE models. We have also used human monoclonal DNRAbs (cloned from peripheral blood B cells of SLE patients), which display reactivity to NMDARs, dsDNA, and DWEYS [45].

When DNRABs bind to the NMDAR, the glutamate-triggered synaptic responses become larger, leading to greater calcium influx into the postsynaptic cell [45] (Fig. 1c). Notably, DNRAB concentrations >100 μg per ml produce NMDAR-dependent responses of such strength that mitochondrial cascades are activated, followed by mitochondrial permeability transition (MPT) pore opening, and apoptosis [39, 45] (Fig. 1d, e). DNRABs can thus be neurotoxic when they are present at sufficient levels in the brain parenchyma and interact with neurons that are enriched with NMDARs. This particular action of DNARBs is comparable to the neural insults that cause neurodegenerative conditions, such as those that occur in Alzheimer and Parkinson diseases. However, it is clear that NPSLE may only parallel a neurodegenerative disorder under severe circumstances, probably when autoantibody is present at high titers in the brain for extended intervals (weeks).

A more likely possibility in NPSLE would be that autoantibodies enter the brain and are extruded rather quickly (hours to days). Indeed, using a mouse model for NPSLE that carries

high levels of circulating DNRABs (termed DNRAB+ mice), we have demonstrated that after the autoantibodies access the brain, their levels remain elevated for 48 h but are undetectable by 14 days [83]. Interestingly, we have shown that antibodies can only enter the brain when DNRAB+ mice are subjected to an insult that breaches their BBB. Even more surprising, the precise nature of the insult determines which brain region exhibits a porous BBB. For instance, treatment with lipopolysaccharide (LPS, a bacterial toxin that produces inflammation) induces a BBB breach around the hippocampus [40], whereas treatment with epinephrine generates a BBB breach in the amygdala [41]. Our in vivo studies in DNRAB+ mice treated with LPS have revealed that they display a clear impairment in spatial cognition when tested in several paradigms, such as the water maze [40], the T-maze for spatial alternation [40, 42], the object-place-memory task [83], and the clock maze [42] (Fig. 1a).

We have conducted in vivo studies in freely moving DNRAB+ mice that are implanted with electrode arrays directed to the CA1 region of the hippocampus in which we record neural signals, such as network oscillations and the activity of place cells [78, 79[79], 85–87]. Our main finding is that place cells become significantly expanded in DNRAB+ mice that have undergone LPS treatment [83] (Fig. 1b). This irregularity becomes evident by ~2 weeks post-LPS treatment, a time when DNRABs are no longer detectable in the hippocampus. These results indicate that the initial DNRAB-mediated action over the NMDARs evolves (after the triggering event is not longer present) and leads to a long-term malfunction in spatial coding that can certainly have a deleterious effect in the spatial cognition of the individual.

The results in the DNRAB+ mouse model strongly suggest that subjects with SLE that are positive for DNRABs (termed DNRAB+ patients) may exhibit noticeable problems with spatial memory. Indeed, a novel neuropsychological task applied to these patients has strengthened this possibility [83]. In the task, the subjects look at drawings of objects (arranged as 2 × 2 arrays) and are later asked either an identification question not addressing spatial relations (recognition memory) or a question about the spatial arrangement of the array (spatial memory). DNRAB+ patients are completely normal in their recognition memory responses but are significantly worse in their spatial memory section when compared to healthy controls [83]. It remains to be demonstrated that the circulating antibodies in DNRAB+ patients actually have traversed the BBB to cause hippocampal deficit, but the data support the hypothesis that poor spatial performance in DNRAB+ patients is attributable to the presence of those antibodies.

In conclusion, the exploration of the effects of DNRABs on NMDARs at various levels of brain organization has generated a multilevel mechanism for their deleterious actions. The animal studies suggest that neurotoxicity may tend to occur in DNRAB+ subjects that exhibit high autoantibody titer and a severely compromised BBB. However, most DNRAB actions may result in destabilization of neural networks, particularly those that underlie spatial coding and spatial cognition if the BBB breach is limited to the hippocampus. We propose that this integrative neuroscience approach provides an invaluable framework to study other poorly understood NPSLE manifestations that are likely caused by brain-permeating autoantibodies.

Acknowledgments

We thank Eric H. Chang, Thomas W. Faust, Sergio Robbiati, Stephen A. Frattini, Toby Klein, and all the members of the LOINN (Laboratory of Immune and Neural Networks) for their help in these studies. This research was funded by NIH Grant P01-AI073693 (to B.D., P.H., M.M., B.V.) and NIH Grant P01-AI102852 (to B.D., P.H., K.T., B.V.).

References

1. Danchenko N, Satia J, Anthony M. Epidemiology of systemic lupus erythematosus: a comparison of worldwide disease burden. Lupus. 2006; 15:308–18. [PubMed: 16761508]
2. Lahita, RG. Systemic lupus erythematosus. 4. Amsterdam: Elsevier; 2004.
3. ACR Ad Hoc Committee on Neuropsychiatric Lupus Nomenclature. The American College of Rheumatology. Nomenclature and case definitions for neuropsychiatric lupus syndromes. Arthritis Rheum. 1999; 42:599–608. [PubMed: 10211873]
4. Brey RL, et al. Neuropsychiatric syndromes in lupus: prevalence using standardized definitions. Neurology. 2002; 58:1214–20. [PubMed: 11971089]
5. Bertsias GK, Boumpas DT. Pathogenesis, diagnosis and management of neuropsychiatric SLE manifestations. Nat Rev Rheumatol. 2010; 6:358–67. [PubMed: 20458332]
6. Ainiala H, et al. The prevalence of neuropsychiatric syndromes in systemic lupus erythematosus. Neurology. 2001; 57:496–500. [PubMed: 11502919]
7. Levy D, Ardoin S, Schanberg L. Neurocognitive impairment in children and adolescents with systemic lupus erythematosus. Nat Clin Pract Rheumatol. 2009; 5:106–14. [PubMed: 19182817]
8. Hanly JG. Neuropsychiatric lupus. Rheum Dis Clin North Am. 2005; 31:273–98. [PubMed: 15922146]
9. Hanly JG, et al. Prospective analysis of neuropsychiatric events in an international disease inception cohort of patients with systemic lupus erythematosus. Ann Rheum Dis. 2010; 69:529–35. [PubMed: 19359262]
10. Sabbadini M, et al. Central nervous system involvement in systemic lupus erythematosus patients without overt neuropsychiatric manifestations. Lupus. 1999; 8:11–9. [PubMed: 10025594]
11. Hanly JG, et al. A prospective analysis of cognitive function and anticardiolipin antibodies in systemic lupus erythematosus. Arthritis Rheum. 1999; 42:728–34. [PubMed: 10211887]
12. Carbotte RM, Denburg SD, Denburg JA. Prevalence of cognitive impairment in systemic lupus erythematosus. J Nerv Ment Dis. 1986; 174:357–64. [PubMed: 3711879]
13. Denburg SD, Carbotte RM, Denburg JA. Cognitive impairment in systemic lupus erythematosus: a neuropsychological study of individual and group deficits. J Clin Exp Neuropsychol. 1987; 9:323–39. [PubMed: 3597726]
14. Hanly JG, Robichaud J, Fisk J. Anti-NR2 glutamate receptor antibodies and cognitive function in systemic lupus erythematosus. J Rheumatol. 2006; 33:1553–8. [PubMed: 16881112]
15. Hanly JG. Diagnosis and management of neuropsychiatric SLE. Nat Rev Rheumatol. 2014; 10:338–47. [PubMed: 24514913]
16. Jeltsch-David H, Muller S. Neuropsychiatric systemic lupus erythematosus: pathogenesis and biomarkers. Nat Rev Neurol. 2014; 10:579–96. [PubMed: 25201240]
17. Olazarán J, et al. Cognitive dysfunction in systemic lupus erythematosus: prevalence and correlates. Eur Neurol. 2009; 62:49–55. [PubMed: 19407455]
18. Kozora E, et al. Inflammatory and hormonal measures predict neuropsychological functioning in systemic lupus erythematosus and rheumatoid arthritis patients. J Int Neuropsychol Soc. 2001; 7:745–54. [PubMed: 11575596]
19. Wallace, DJ.; Hahn, B.; Dubois, EL. Dubois' lupus erythematosus. 5. Baltimore: Williams & Wilkins; 1997.
20. Carbotte RM, Denburg SD, Denburg JA. Cognitive dysfunction in systemic lupus erythematosus is independent of active disease. J Rheumatol. 1995; 22:863–7. [PubMed: 8587073]
21. McLaurin EY, et al. Predictors of cognitive dysfunction in patients with systemic lupus erythematosus. Neurology. 2005; 64:297–303. [PubMed: 15668428]

22. Harboe E, et al. Neuropsychiatric syndromes in patients with systemic lupus erythematosus and primary Sjögren syndrome: a comparative population-based study. Ann Rheum Dis. 2009; 68:1541–6. [PubMed: 18930990]
23. Utset TO, et al. Prevalence of neurocognitive dysfunction and other clinical manifestations in disabled patients with systemic lupus erythematosus. J Rheumatol. 2006; 33:531–8. [PubMed: 16511923]
24. Appenzeller S, et al. Cognitive impairment and employment status in systemic lupus erythematosus: a prospective longitudinal study. Arthritis Rheum. 2009; 61:680–7. [PubMed: 19405004]
25. Roebuck-Spencer TM, et al. Use of computerized assessment to predict neuropsychological functioning and emotional distress in patients with systemic lupus erythematosus. Arthritis Rheum. 2006; 55:434–41. [PubMed: 16739211]
26. Kozora E, et al. Cognitive dysfunction in systemic lupus erythematosus: past, present, and future. Arthritis Rheum. 2008; 58:3286–98. [PubMed: 18975345]
27. Petri M, et al. Cognitive function in a systemic lupus erythematosus inception cohort. J Rheumatol. 2008; 35:1776–81. [PubMed: 18634154]
28. Bertsias GK, et al. EULAR recommendations for the management of systemic lupus erythematosus with neuropsychiatric manifestations: report of a task force of the EULAR standing committee for clinical affairs. Ann Rheum Dis. 2010; 69:2074–82. [PubMed: 20724309]
29. Brunner HI, et al. Initial validation of the Pediatric Automated Neuropsychological Assessment Metrics for childhood-onset systemic lupus erythematosus. Arthritis Rheum. 2007; 57:1174–82. [PubMed: 17907235]
30. Cieślik P, et al. Vasculopathy and vasculitis in systemic lupus erythematosus. Pol Arch Med Wewn. 2008; 118:57–63. [PubMed: 18405174]
31. Fietta P, et al. Psychiatric and neuropsychological manifestations of systemic lupus erythematosus. Acta Biomed. 2011; 82:97–114. [PubMed: 22480065]
32. Golan TD. Lupus vasculitis: differential diagnosis with antiphospholipid syndrome. Curr Rheumatol Rep. 2002; 4:18–24. [PubMed: 11798978]
33. Bluestein HG, Zvaifler NJ. Brain-reactive lymphocytotoxic antibodies in the serum of patients with systemic lupus erythematosus. J Clin Invest. 1976; 57:509–16. [PubMed: 56343]
34. Bresnihan B, et al. Brain reactivity of lymphocytotoxic antibodies in systemic lupus erythematosus with and without cerebral involvement. Clin Exp Immunol. 1977; 30:333–7. [PubMed: 606449]
35. Bresnihan B, et al. An antineuronal antibody cross-reacting with erythrocytes and lymphocytes in systemic lupus erythematosus. Arthritis Rheum. 1979; 22:313–20. [PubMed: 371627]
36. How A, et al. Antineuronal antibodies in neuropsychiatric systemic lupus erythematosus. Arthritis Rheum. 1985; 28:789–95. [PubMed: 4015726]
37. Temesvari P, et al. Serum lymphocytotoxic antibodies in neuropsychiatric lupus: a serial study. Clin Immunol Immunopathol. 1983; 28:243–51. [PubMed: 6872362]
38. Wilson HA, et al. Association of IgG anti-brain antibodies with central nervous system dysfunction in systemic lupus erythematosus. Arthritis Rheum. 1979; 22:458–62. [PubMed: 375939]
39. DeGiorgio LA, et al. A subset of lupus anti-DNA antibodies cross-reacts with the NR2 glutamate receptor in systemic lupus erythematosus. Nat Med. 2001; 7:1189–93. [PubMed: 11689882]
40. Kowal C, et al. Cognition and immunity; antibody impairs memory. Immunity. 2004; 21:179–88. [PubMed: 15308099]
41. Huerta PT, et al. Immunity and behavior: antibodies alter emotion. Proc Natl Acad Sci USA. 2006; 103:678–83. [PubMed: 16407105]
42. Kowal C, et al. Human lupus autoantibodies against NMDA receptors mediate cognitive impairment. Proc Natl Acad Sci USA. 2006; 103:19854–9. [PubMed: 17170137]
43. Lee JY, et al. Neurotoxic autoantibodies mediate congenital cortical impairment of offspring in maternal lupus. Nat Med. 2009; 15:91–6. [PubMed: 19079257]
44. Diamond B, et al. Losing your nerves? Maybe it's the antibodies. Nat Rev Immunol. 2009; 9:449–56. [PubMed: 19424277]

45. Faust TW, et al. Neurotoxic lupus autoantibodies alter brain function through two distinct mechanisms. Proc Natl Acad Sci USA. 2010; 107:18569–74. [PubMed: 20921396]
46. Wang L, et al. Female mouse fetal loss mediated by maternal autoantibody. J Exp Med. 2012; 209:1083–9. [PubMed: 22565825]
47. Bloom O, et al. Generation of a unique small peptidomimetic that neutralizes lupus autoantibody activity. Proc Natl Acad Sci USA. 2011; 108:10255–9. [PubMed: 21646518]
48. Diamond B, et al. Moving towards a cure: blocking pathogenic antibodies in systemic lupus erythematosus. J Intern Med. 2011; 269:36–44. [PubMed: 21158976]
49. Mackay M, et al. Differences in regional brain activation patterns assessed by functional magnetic resonance imaging in patients with systemic lupus erythematosus stratified by disease duration. Mol Med. 2011; 17:1349–56. [PubMed: 21953419]
50. Fragoso-Loyo H, et al. Serum and cerebrospinal fluid autoantibodies in patients with neuropsychiatric lupus erythematosus. Implications for diagnosis and pathogenesis. PLoS ONE. 2008; 3:e3347. [PubMed: 18836530]
51. Yoshio T, et al. Association of IgG anti-NR2 glutamate receptor antibodies in cerebrospinal fluid with neuropsychiatric systemic lupus erythematosus. Arthritis Rheum. 2006; 54:675–8. [PubMed: 16447246]
52. Steup-Beekman G, et al. Anti-NMDA receptor autoantibodies in patients with systemic lupus erythematosus and their first-degree relatives. Lupus. 2007; 16:329–34. [PubMed: 17576734]
53. Arinuma Y, et al. Association of cerebrospinal fluid anti-NR2 glutamate receptor antibodies with diffuse neuropsychiatric systemic lupus erythematosus. Arthritis Rheum. 2008; 58:1130–5. [PubMed: 18383393]
54. Bosch X, et al. The DWEYS peptide in systemic lupus erythematosus. Trends Mol Med. 2012; 18:215–23. [PubMed: 22365619]
55. Lauvsnes MB, Omdal R. Systemic lupus erythematosus, the brain, and anti-NR2 antibodies. J Neurol. 2012; 259:622–9. [PubMed: 21909801]
56. Matus S, et al. Antiribosomal-P autoantibodies from psychiatric lupus target a novel neuronal surface protein causing calcium influx and apoptosis. J Exp Med. 2007; 204:3221–34. [PubMed: 18056288]
57. Kivity S, et al. Abnormal olfactory function demonstrated by manganese-enhanced MRI in mice with experimental neuropsychiatric lupus. Ann N Y Acad Sci. 2010; 1193:70–7. [PubMed: 20398010]
58. Jacob A, et al. C5a alters blood-brain barrier integrity in experimental lupus. FASEB J. 2010; 24:1682–8. [PubMed: 20065106]
59. Sakić B. The MRL model: an invaluable tool in studies of autoimmunity-brain interactions. Methods Mol Biol. 2012; 934:277–99. [PubMed: 22933151]
60. Gielen M, et al. Mechanism of differential control of NMDA receptor activity by NR2 subunits. Nature. 2009; 459:703–7. [PubMed: 19404260]
61. Collingridge GL, Kehl SJ, McLennan H. Excitatory amino acids in synaptic transmission in the Schaffer collateral-commissural pathway of the rat hippocampus. J Physiol. 1983; 334:33–46. [PubMed: 6306230]
62. Ozawa S, Kamiya H, Tsuzuki K. Glutamate receptors in the mammalian central nervous system. Prog Neurobiol. 1998; 54:581–618. [PubMed: 9550192]
63. Huntley GW, Vickers JC, Morrison JH. Cellular and synaptic localization of NMDA and non-NMDA receptor subunits in neocortex: organizational features related to cortical circuitry, function and disease. Trends Neurosci. 1994; 17:536–43. [PubMed: 7532339]
64. Wenthold RJ, et al. Evidence for multiple AMPA receptor complexes in hippocampal CA1/CA2 neurons. J Neurosci. 1996; 16:1982–9. [PubMed: 8604042]
65. Regalado MP, Villarroel A, Lerma J. Intersubunit cooperativity in the NMDA receptor. Neuron. 2001; 32:1085–96. [PubMed: 11754839]
66. Kutsuwada T, et al. Molecular diversity of the NMDA receptor channel. Nature. 1992; 358:36–41. [PubMed: 1377365]

67. Collingridge GL, Singer W. Excitatory amino acid receptors and synaptic plasticity. Trends Pharmacol Sci. 1990; 11:290–6. [PubMed: 2167544]
68. Coan EJ, Collingridge GL. Magnesium ions block an N-methyl-D-aspartate receptor-mediated component of synaptic transmission in rat hippocampus. Neurosci Lett. 1985; 53:21–6. [PubMed: 2859558]
69. Aarts M, et al. Treatment of ischemic brain damage by perturbing NMDA receptor- PSD-95 protein interactions. Science. 2002; 298:846–50. [PubMed: 12399596]
70. Aarts M, Tymianski M. Molecular mechanisms underlying specificity of excitotoxic signaling in neurons. Curr Mol Med. 2004; 4:137–47. [PubMed: 15032710]
71. Hong SJ, Dawson TM, Dawson VL. Nuclear and mitochondrial conversations in cell death: PARP-1 and AIF signaling. Trends Pharmacol Sci. 2004; 25:259–64. [PubMed: 15120492]
72. Cull-Candy SG, Leszkiewicz DN. Role of distinct NMDA receptor subtypes at central synapses. Science. 2004; 255:16–25.
73. Cajal, SR. Histologie du systeme nerveux de l'homme et des vertebrae. Paris: Moline; 1911.
74. Amaral, DG.; Witter, MP. Hippocampal formation. In: Paxinos, G., editor. The rat nervous system. New York: Academic Press; 1995. p. 443-93.
75. Scoville WB, Milner B. Loss of recent memory after bilateral hippocampal lesions. J Neurochem. 1957; 20:11–21.
76. Mishkin M, Vargha-Khadem F, Gadian DG. Amnesia and the organization of the hippocampal system. Hippocampus. 1998; 8:212–6. [PubMed: 9662136]
77. Morris, RGM. Theories of hippocampal function. In: Andersen, P.; Morris, RM.; Amaral, D.; Bliss, T.; O'Keefe, J., editors. The Hippocampus Book. New York: Oxford University Press; 2007. p. 581-714.
78. O'Keefe, J.; Nadel, L. The hippocampus as a cognitive map. New York: Oxford University Press; 1978.
79. O'Keefe, J. Hippocampal neurophysiology in the behaving animal. In: Morris, RM.; Amaral, D.; Bliss, T.; O'Keefe, J., editors. The Hippocampus Book Andersen P. New York: Oxford University Press; 2007. p. 475-548.
80. Morris RG, et al. Place navigation impaired in rats with hippocampal lesions. Nature. 1982; 297:681–3. [PubMed: 7088155]
81. Morris RG, et al. Selective impairment of learning and blockade of long-term potentiation by an N-methyl-D-aspartate receptor antagonist, AP5. Nature. 1986; 319:774–6. [PubMed: 2869411]
82. McEchron MD, et al. Hippocampectomy disrupts auditory trace fear conditioning and contextual fear conditioning in the rat. Hippocampus. 1998; 8:638–46. [PubMed: 9882021]
83. Tsien JZ, Huerta PT, Tonegawa S. The essential role of hippocampal CA1 NMDA receptor-dependent synaptic plasticity in spatial memory. Cell. 1996; 87:1327–38. [PubMed: 8980238]
84. Huerta PT, et al. Formation of temporal memory requires NMDA receptors within CA1 pyramidal neurons. Neuron. 2000; 25:473–80. [PubMed: 10719900]
85. Chang EH, et al. Selective impairment of spatial cognition caused by autoantibodies to the *N*-methyl-d-aspartate receptor. EBio-Medicine. 2015; 2:755–64.
86. Chang EH, Frattini SA, Robbiati S, Huerta PT. Construction of microdrive arrays for chronic neural recordings in awake behaving mice. J Vis Exp. 2013; 77:e50470.doi: 10.3791/50470 [PubMed: 23851569]
87. Faust TW, Robbiati S, Huerta TS, Huerta PT. Dynamic NMDAR-mediated properties of place cells during the object place memory task. Front Behav Neurosci. 2013; 7:202. [PubMed: 24381547]

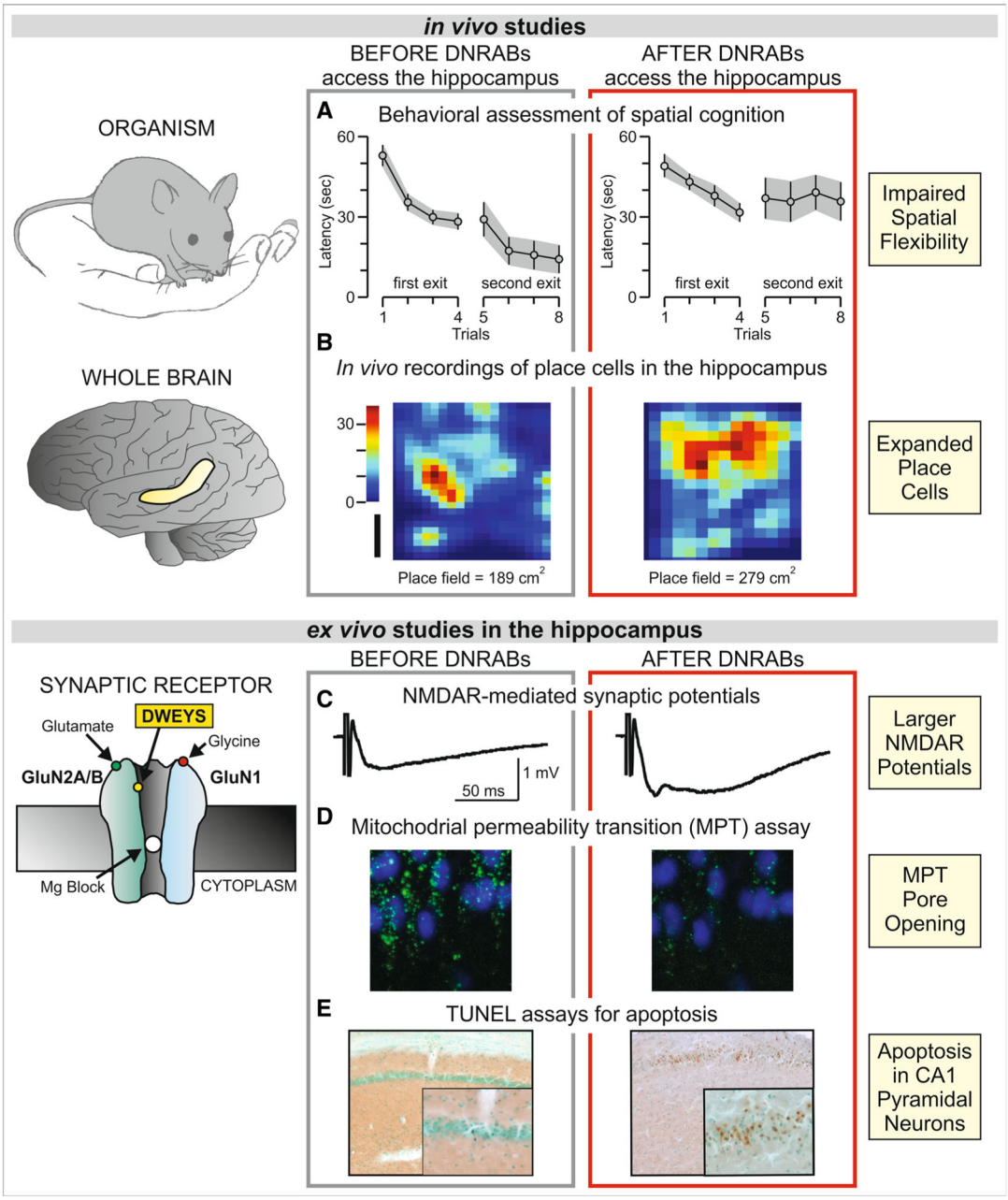

Fig. 1.

Analysis of the effects of DNRABs at different levels of neural organization. **a** The graphs show DNRAB+ mice assessed in the clock maze, a test for spatial cognition [42]. For this task, the apparatus is a circular base platform (diameter, 85 cm) attached to a waterproof clear wall (30 cm high). Cold water (20 °C) is added to a depth of 2 cm. The perimeter wall is pierced by 12 holes, 4 cm in diameter, arranged equidistantly around the circumference so that they are 23 cm apart, like the 12 h on a clock face. The lower edge of each hole is 3 cm above the maze floor, that is, at mouse head level. Eleven of these tubes are sealed with black plugs, flush with the internal pool wall surface; one is open and leads to an escape

pipe, which is 4 cm in diameter, made of black flexible plastic. Thus, from within the clock maze, the true exit looks similar to the decoys, even to the human eye. The pool is surrounded by distal unmoving cues, which are illuminated by focal white lights within a darkened testing room. The task consists in escaping into the open tube and connected pipe; the pipe is then removed and the mouse is transported to the nearby home cage. On the first day, the mice are trained to the same exit for four trials, while on the next day a different exit is used. *Left*, this group ($n = 10$) has circulating DNRABs that do not enter the brain and shows normal acquisition. *Right*, this group ($n = 10$) is tested 4 weeks post-LPS (which allows DNRAB entry into the hippocampus) and shows significantly longer latency to find a second exit ($P < 0.01$, ANOVA for trials 5–8), a deficit in spatial flexibility. **b** *Left*, side view of the brain with the hippocampus highlighted in yellow. *Right*, neural recordings in freely moving animals show that DNRAB+ mice exhibit larger place fields after DNRABs enter this brain region. For these experiments, mice ($n = 8$) are implanted with multielectrode arrays (4 tetrodes), directed to dorsal CA1 (coordinates of −2.0 mm AP, +1.6 mm ML from bregma) [83–85]. Neural activity is recorded with a unitary gain headstage preamplifier (HS-18; Neuralynx Bozeman, MT) that is connected to a programmable amplifier (Lynx-8, Neuralynx) linked to Cheetah-32 software (Neuralynx), which acquires single units at a sampling rate of 30 kHz (band-pass filter, 600–6 kHz). The headstage also includes two LEDs that are used for tracking the animal's position at 30 frames per second, which is accomplished by linking an infrared-sensitive camera, mounted above the chamber, to the video input of the Cheetah software. Three days post-surgery, the implanted mice are placed in the behavioral arena over a period of 3–5 days. The figure shows representative firing rate maps, recorded 2 weeks pre-LPS and 4 weeks post-LPS, during 10-min sessions in an arena (viewed from the top, 40 cm on the side). *Color scale* indicates frequency (Hz, spikes per second), in which *red* corresponds to the peak firing rate and *blue* to null firing. The size of the place field is indicated below the arena. **c** *Left*, cartoon of the NMDAR showing the binding sites for glutamate and other ligands, as well as the DWEYS consensus sequence. *Right*, studies in *ex vivo* hippocampal slices reveal that application of DNRABs significantly enhances the size of the extracellularly recorded NMDAR-mediated synaptic potentials. **d–e** At high titer of DNRABs (>100 μg per ml), hippocampal pyramidal neurons undergo MPT pore opening (**d**, detected as loss of *green* fluorescence in the *right panel*; the *blue color* represents staining of cell bodies, 100 μm on the side), and apoptosis (**e**, detected as *brown staining* by TUNEL assay, especially noticeable in the *right panel inset*)