Clinical presentations and antibody mechanisms in anti-IgLON5 disease

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Abstract

Anti-IgLON5 disease is a rare neurological disease, identified just ten years ago, where autoimmunity and neurodegeneration converge. The heterogeneity of symptoms, sometimes mimicking pure neurodegenerative diseases or motor neuron diseases, in addition to lack of awareness, represents a diagnostic challenge. Biomarkers of neuronal damage in combination with in vivo visualization of tau deposition using positron emission tomography (PET) scanning could represent a major advance in monitoring disease progression. Recent studies with more autopsies available have helped refine the knowledge of the pathological features of the disease and strengthen the autoimmune hypothesis of the disease. Although the pathogenesis of anti-IgLON5 disease remains unclear, the irreversible antibody-mediated decrease of IgLON5 clusters from the cell surface and alterations produced in the cytoskeleton, as well as the behavioural abnormalities and signs of neuroinflammation and neurodegeneration observed in the brains of animals infused with antibodies from patients by passive transfer, which have recently been published, support the autoimmune hypothesis of the disease. This review aims to summarize these important aspects and recent advances in the pathophysiology of anti-IgLON5 disease.

Keywords: Anti-IgLON5 disease, autoantibodies, neurodegeneration, biomarkers, animal models.

1. Introduction

Anti-IgLON5 disease is a rare neurological disease (Orpha: 420789) where autoimmunity and neurodegeneration converge. Ten years ago, we identified a group of patients with a distinctive and prominent sleep disorder, gait instability and bulbar symptoms bearing an unknown autoantibody against the neuronal cell surface in serum and CSF [1]. IgLON5 was unequivocally revealed as the target of this new reactivity but the physiological function of this immunoglobulin-like cell adhesion molecule belonging to the IgLON family of proteins was unexplored. Unlike other autoimmune encephalitis [2], progression of the disease was mainly chronic and unresponsive to immunotherapy; moreover, neuropathological examination of the few available autopsies revealed the unexpected discovery of an atypical tauopathy mainly restricted to neurons in hypothalamic and brainstem areas [1,3]. Therefore, the intriguing question arose whether the observed tau pathology was a consequence of IgLON5 antibodies or, conversely, the presence of these antibodies was a mere secondary event of the tauopathy. The research conducted during these years since its discovery has helped to define and broaden the clinical phenotype of the disease, and to gain insights into the mechanisms of the antibodies against IgLON5, which support the autoimmune hypothesis of the pathogenesis of anti-IgLON5 disease.

2. Epidemiology

At present, there are no studies that address the prevalence and incidence of anti-IgLON5 disease, but although we can presume that it is going to be low, according to our experience as reference center for the diagnostic of autoimmune related-CNS disorders, it could be the third most common after NMDAR and LGI1 encephalitides. From 2017 to 2021, we identified 240 patients with antibodies against cell surface

antigens, finding IgLON5 antibodies in 10% of them (unpublished data). Patients with anti-IgLON5 disease are typically elderly adults, being the median age at diagnosis in our cohort of 94 patients around 67 years (range: 46-91 years) with a slightly predominance of males (56.4%). Heterogeneity of symptoms and lack of awareness about diagnostic red flags contribute to the under diagnosis of the disease.

3. Diagnosis

3.1 Clinical phenotypes and mimics

The clinical manifestations of anti-IgLON5 disease are heterogeneous involving multiple levels of the central nervous system, thus representing a diagnosis challenge. Most patients present a combination of symptoms including a distinctive sleep disorder with NREM and REM parasomnias with stridor, obstructive sleep apnoea, gait instability, abnormal movements (chorea and craniofacial dyskinesia), and bulbar symptoms like dysphagia and dysarthria [4–6]. The progression of the disease is mainly chronic or insidious over months or years which differ from the typical subacute presentation of other known autoimmune neurological disorders associated to antibodies targeting cell surface antigens [2]. The few cases with anti-IgLON5 disease (25-28%) that follow a subacute clinical course would be more rapidly diagnosed according to an observational retrospective study in 53 patients [7], but the most frequent scenario is a prolonged diagnostic delay of around 2-3 years, after consulting with up to 4 different physicians, and when the patient already needs assistance for daily activities and ranking 3 in the modified Rankin Score (mRS) [7]. This is a crucial point that should be improved in the future by promoting awareness of the disease and dissemination of the expanding clinical phenotypes attributed to anti-IgLON5 disease. Although a great overlapping of symptoms can be found in most of the patients, the following clinical

manifestations (in order of frequency) would indicate searching for the presence of IgLON5 antibodies (Figure 1) and the possible mimics will be also reviewed (Figure 2).

3.1.1 Bulbar syndrome with prominent dysphagia, dysarthria, with episodes of respiratory failure due to central hypoventilation or obstruction secondary to vocal cord palsy

This is the most frequent phenotype (around 40% of cases) at onset. Bulbar dysfunction is indeed present at the time of diagnosis in most of the patients with anti-IgLON5 disease (74-90%) manifested as mild and intermittent dysphagia, dysarthria, laryngeal stridor or episodes of respiratory failure [6,8]. Dysphagia at disease onset can be moderate or severe in up 36% of the patients [7,9]; in this case enteral nutrition or a feeding tube may be necessary due to the subsequent weight loss. Dysarthria presenting with dysphonia, hoarseness or slurred speech is also frequent, but it is usually mild [10]. Laryngoscopy shows mild to moderate vocal cord palsy in around 50-60% of the patients which can cause the stridor observed during sleep [6]. The more severe complications are episodes of central hypoventilation or laryngeal spams leading to admission at the UCI and requiring ventilatory support or a permanent tracheostomy afterwards. This phenotype can mimic myasthenia gravis or motor neuron disease if apart from prominent dysphagia patients present with fasciculations or blepharoptosis [[9,11] (see also neuromuscular phenotype).

3.1.2 Gait instability, abnormal movements or PSP-like syndrome

Gait and movement disorders occur in 87% of patients with anti-IgLON5 disease and frequently are a common reason for initial neurologic consultation in around a half of them. Gait impairment, often associated with recurrent falls, is present in 72% of the patients [12]. Disequilibrium and alteration of postural reflexes are the main cause of frequent falls or gait impairment, but cerebellar ataxia also occurs in some patients

[1,6,13,14]. Generalized chorea and craniofacial dyskinesias (e.g dystonia, myorhythmia or myokymia) are present in up to one third of the patients [15–17]. In some patients, Huntington's disease can be suspected if chorea is associated with cognitive impairment. Other movements disorders less frequently reported are abdominal dyskinesias, spams in lower limbs or parkinsonism, often combined with gait instability. Abnormal eye movements are observed in 10-20% of patients with gait disorder; thus, patients with this phenotype may be misdiagnosed with progressive supranuclear palsy (PSP) [15,18,19]. However, very few patients fulfilling PSP criteria are positive for IgLON5 antibodies and the concomitance of other neurological symptoms such as prominent sleep dysfunction, stridor and respiratory problems can ultimately lead to the correct diagnostic of IgLON5 disease. Furthermore, dissimilar to PSP, patients with anti-IgLON5 disease present horizontal or upward gaze palsy with preservation of downward movements [6,18,19].

3.1.3 Sleep problems with NREM and REM abnormal movements and behaviors, obstructive sleep apnea and stridor

The sleep problems of these patients are so distinctive and unique that promoted the identification of the disease [1]. The complex features of the sleep disorder which is present in almost 90% of the patients are characterized by a distinctive non-REM sleep parasomnia, REM behaviour disorder (RBD), obstructive sleep apnoea (OSA) and stridor. Video-polysomnography studies evidence parasomnias at sleep initiation with undifferentiated NREM sleep and poorly structured N2 NREM sleep that associate with electromyographic activation and vocalizations, simple or finalistic movements resembling daytime activities. Patients with anti-IgLON5 disease, differently from *agrypnia excitata* where sleep is severely reduced or even absent, usually show a progressive normalization of N3 sleep periods, especially at the end of the night with

frequent spindles, K complexes, and delta slowing, without electromyographic activation. RBD manifests with multifocal jerking and excessive tonic and phasic muscle activity. Breathing problems during sleep are also common and include OSA and stridor, which can be ameliorated by continuous positive air pressure (CPAP) therapy [4,10]. Parasomnia can be difficult to treat. The sleep disorder may be absent at onset in 10-20% of the cases [4,5,20], overlooked if neurologists do not specifically inquire, overshadowed by more severe symptoms or even appear later in the course of the disease.

3.1.4 Cognitive impairment, psychiatric manifestations and behavioural changes

Although cognitive impairment is present in around 30% of the cases [7], very few studies have addressed the neuropsychological assessment of these patients in whom deficits of concentration, impaired verbal and visual memory, and difficulties in executive tasks have been observed [21]. Furthermore, some patients fulfil dementia criteria. A recent study in a large cohort of 920 patients with neurodegenerative dementia found IgLON5 antibodies in 3 of them [22]. Awareness of this possibility is necessary to differentiate pure neurodegenerative disorders from autoimmune-mediated syndromes that potentially could benefit from immunotherapy. Hallucinations, psychosis, aggressive behaviours or other psychiatric manifestations are less common.

3.1.5 Neuromuscular syndrome including muscle hyperexcitability with stiffness and cramps, limb weakness, muscle wasting, or fasciculation

The complex and heterogeneous constellation of clinical symptoms of anti-IgLON5 disease include also neuromuscular manifestations and although muscle fasciculations are clinically evident only in 10-19% of the patients, they are detected by electromyography in around 34% [7,8]. Importantly, anti-IgLON5 disease can mimic myasthenic syndromes when mild to moderated proximal progressive muscle weakness

combined with dysphagia or eyelid ptosis are present. Bulbar dysfunction, muscle fasciculation, atrophy and weakness may also suggest the diagnosis of motor neuron disease or might mimic stiff-person syndrome if the patient additionally shows symptoms of nervous system hyperexcitability (e.g exaggerated startle, stiffness, spasm, or cramps) [11,23,24].

3.2 Paraclinical studies, biomarkers and prognostic scales

The diagnosis of anti-IgLON5 disease is based on the detection of IgLON5 antibodies in serum or CSF by immunohistochemistry on rat brain tissue (Figure 3) and confirmation by a specific cell-based assay with HEK cells transfected with IgLON5 plasmid (Figure 4). All the patients have antibodies in the serum, and 90% of them are also detected in the CSF. Other paraclinical studies are negative or non-informative in most cases. Interestingly, brain MRI which is usually normal or only displays a mild atrophy [25], showed more inflammatory evidence in patients with a subacute presentation [26]. Furthermore, pleocytosis is more frequently observed in patients with a closest CSF collection to the onset of the disease. However, CSF pleocytosis (<10 lymphocytes/ul) and increase of proteins are mild. About 60% of patients harbour the HLA class II haplotype DRB1*10:01/DQB1*05:01 (DR10/DQ5), which is infrequent in the normal population (1-3%) [27]. This robust association favours the hypothesis of the autoimmune origin of the disease and interestingly the presence of these alleles correlates with the bulbar dysfunction in one study and with the typical sleep disorder in two [7,27]. In addition, patients without the DR10/DQ5 haplotype are more frequently positive for IgLON5-abs only in the serum and have a PSP-like phenotype [27].

Recently, a Genome-wide association study in a multicentric study including 62 patients identified also the association of the disease with the HLA locus but results pointed out to a major relevance of DQ5 which is in strong linkage disequilibrium with DR10 [28].

Importantly, a recent study reports that tau deposition can be *in vivo* visualized by the tau PET scan tracer [18F]PI-2620 in patients with anti-IgLON5 disease and could even correlate with the extent of the disease [29]. Although the cohort of 4 patients is small and more studies are needed to validate the results, if confirmed it could be useful for the neuroimaging assessment of response to therapy. Another promising biomarker is the measurement of neurofilament light chain levels (NfL) in serum. An observational retrospective study in 53 patients found that high serum levels of NfL at diagnostic would be indicative of poor prognosis [7]. Another important advance in the management and criteria of the disease has been the establishment of a clinical scale, the IgLON5 composite score (ICS), to assess the severity of the disease by weighing the symptoms by clinical domains [30]. This could be useful to measure the evolution of the disease or the response to immunotherapy in a uniform way between centers. (Table 1)

3.3 Recent advances in neuropathological criteria of anti-IgLON5 disease

The neuropathological examinations of the first two available autopsies revealed the presence of an atypical tauopathy mainly restricted to neurons involving the hypothalamus and the tegmentum of the brainstem. Neuronal loss, astrogliosis and microglial activation were observed in these brain areas. Tau aggregates were detected only in neurons and presence of three-repeat (3R) and four-repeat (4R) tau isoforms were confirmed by immunohistochemistry but inflammatory infiltrates or other deposits like beta-amyloid were absent [1,3]. In a posterior study, the previously identified brainstem-predominant tauopathy was not detected in a new case with anti-IgLON5 disease but only phospho-tau deposits restricted to medial-temporal regions compatible with age-related comorbidity. Interestingly, a biochemical assay aimed at investigating the electrophoretic bar-code of the insoluble tau aggregates differentiated this patient that showed a similar pTau band pattern with Alzheimer's disease from a previous brainstem-predominant patient that presented a

distinctive band pattern signature [31] (Figure 5). Post-mortem diagnostic criteria were defined thereafter when more autopsies were available. All the brains showed the hypothalamic/brainstem tauopathy with a cranio-caudal gradient of severity until the upper cervical cord and the main features found coincided with the ones already reported [3] (Figure 6). However, a relevant recent neuropathological study on 9 cases shed light about the pathophysiology of the disease suggesting that the tau pathology could be a late event in the disease after an initial inflammatory process. Tauopathy was detected in 6 patients with a prolonged evolution of the disease (median 9 years). Five of them had the described brainstem neuronal tauopathy and one fulfilled neuropathological criteria for progressive supranuclear palsy (PSP) with a prominent neuronal and glial 4R tauopathy. Interestingly, three patients with a relatively short duration (median 1.25 years) showed only a primary age-related neurofibrillary pathology. Inflammatory infiltrates of B and T cells were found independently of the presence of tauopathy. Furthermore, substantial deposition of IgG4 antibodies were found especially in two cases without tauopathy and short duration of the disease in brain areas relevant for the disease such as brainstem tegmentum that might be prone to develop subsequently tau pathology, although the exact role of IgLON5 antibodies of IgG4 subclass has not been fully elucidated yet [32] (Figure 7). Overall, these new results support the autoimmune hypothesis of the pathogenesis of the disease.

4. Mechanisms of IgLON5 antibodies and animal models

The hallmark of the disease is the presence of antibodies against IgLON5 but the physiological function of this cell surface adhesion protein is poorly understood. IgLON5 is a glycosylphosphatidylinositol (GPI)-anchored protein to the plasma membrane with three extracellular immunoglobulin-like (Ig) domains [33]. The immunodominant region is in the second Ig domain, and although IgLON5-abs are IgG4 subclass predominantly, most samples also have some content of IgG1. In vitro

studies with rat hippocampal neurons demonstrated that the IgLON5-IgG1 fraction internalizes the antigen after crosslinking, providing clues to the pathogenicity of IgLON5-abs [33]. Furthermore, total purified IgG from samples with either predominant IgG4 or IgG1 IgLON5-abs indistinctly induced cytoskeletal abnormalities with the presence of swellings, bulb-like, ring structures and premature termination of dendrites [34] (Figure 8). However, the affected pathways leading to this effect, which could be considered an early neurodegeneration event and the connection between a non-transmembrane protein and the cytoskeleton, are unknown. Moreover, development of hyperphosphorylated tau deposits as consequence of the autoantibodies' effects has been explored in one study using derived neurons from IPSCs and purified IgG of a single case, but this needs further confirmation [35].

IgG4 antibodies poorly activate complement by the classical pathway and have the ability to perform Fab arm exchange [36] which prevents the crosslinking and internalization of the target antigen; therefore, the most likely pathogenic mechanism exerted by IgG4 autoantibodies is the loss of function of the targeted antigen by interfering in the interaction with its binding partners [37]. In the case of IgLON5, IgLON5-abs pulled-down peptides of other members of IgLON family from rat cerebellar neurons when analyzed by mass spectrometry studies, revealing that IgLONs interact between them. Furthermore, IgLON5 is spontaneously shed to the medium of cultured neurons and transfected HEK cells with human IgLON5 plasmid. We demonstrated that IgLON5-abs interfered with the interactions of this soluble IgLON5 with the other IgLONs, but this potentially pathogenic mechanism was independent of the IgG subclass, at least when assayed in vitro [38] (Figure 9).

However, the very few published in vivo models of passive transfer of the disease have not fully reproduced the clinical symptoms of the patients. First, a pilot study injected

through osmotic pumps purified IgLON5-IgG and control IgG into the ventricular compartment of WT mice or congenic mice expressing human tau. This animal model of passive transfer of IgLON5 antibodies aimed to study whether the antibodies could reproduce the sleep disorder of the patients but was basically negative and animals did not show major behavioral abnormalities. Interestingly, the analysis of the brains showed substantial more tau pathology in female mice that received IgLON5 antibodies [39]. A second study followed two experimental approaches for the passive transfer of the antibodies to the brain of the animals: 1) continuous infusion into the lateral ventricle, and 2) parenchymal injection into the CA1 layer of the hippocampus of IgLON5-IgG or IgG control [40]. Animals receiving IgLON5-IgG showed long term memory deficits in the two experimental designs plus anxiety-like behavior when the IgG was intraventricular injected. Signs of inflammation and persistent deposition of human IgG was also observed. In another study, mice injected with IgLON5-IgG in the substantia nigra pars compacta showed motor balance impairment, dopamine decrease, inflammation and a moderate increase of phospho-tau three months post-injection [41]. Altogether these results are in line with the autoimmune hypothesis of the pathogenesis of the disease.

Moreover, subtle behavioral deficits were observed in knockout mice when IgLON5 gene was silenced by CRISPR-Cas9 (IgLON5-KO) in two independent studies. In the first study, IgLON5-KO males had more slips in the beam balance test, which assesses motor and vestibular function by quantifying the ability to balance on a narrow wooden beam. Although statistical analyses did not reach significance in the female IgLON5-KOs, they showed the same trend as in males. These results suggest that IgLON5 has a physiological role in fine motor coordination and balance; these findings support the possibility that IgLON5 autoantibodies can interfere with IgLON5's function and may

be pathogenically involved in the gait instability frequently noted in this disorder. An independent study from another group has recently confirmed these results demonstrating subtle behavioral abnormalities including motor dysfunction in other IgLON5-KO mice [42]. This is also consistent with the results of the passive transfer of IgLON5 antibodies into the nigrostriatal dopaminergic pathway of mice which produced sustained motor impairment [41]. More studies are needed in the future to understand the link between IgLON5 antibodies and tau pathological events.

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Figure legends

Figure 1: Circular graph representing in percentage the most abundant clinical phenotypes of the anti-IgLON5 disease at diagnosis based on a series of 86 patients.

Figure 2: Diagram showing the most frequent clinical symptoms of anti-IgLON5 disease in combination with subtypes and possible mimics according to overlapping symptoms. *Taken from Gaig C, Sabater L. New knowledge on anti-IgLON5 disease. Curr Opin Neurol. 2024 Jun 1;37(3):316-321. doi: 10.1097/WCO.0000000000001271. Epub 2024 Apr 1. PMID: 38563128; PMCID: PMC11064895.*

Figure 3: Anti-IgLON5 reactivity demonstrated by immunohistochemistry on rat brain tissue and by immunofluorescence performed on live hippocampal neurons. A) Typical diffuse neuropil staining of IgLON5 antibodies from the CSF of a patient on sagittal sections of rat brain tissue. B) CSF from an Alzheimer disease patient served as negative control. C) Magnification of the cerebellum showing the diffuse staining of the molecular cell layer and the synaptic staining of the glomerula in the granular cell layer. D) Magnification of the Purkinje cell layer of the cerebellum, counterstained with hematoxylin). E) Intense staining of the cell surface of rat hippocampal neurons when incubated live with a patient's serum. Scale bars in A and B=1000 μm, C=200 μm, D=50 μm and E=20 μm. *Taken from Sabater L, Gaig C, Gelpi E, Bataller L, Lewerenz J, Torres-Vega E, Contreras A, Giometto B, Compta Y, Embid C, Vilaseca I, Iranzo A, Santamaría J, Dalmau J, Graus F. A novel non-rapid-eye movement and rapid-eye-movement parasomnia with sleep breathing disorder associated with antibodies to IgLON5: a case series, characterisation of the antigen, and post-mortem study. Lancet Neurol. 2014 Jun;13(6):575-86. doi: 10.1016/S1474-4422(14)70051-1. Epub 2014 Apr 3. Erratum in: Lancet Neurol. 2015 Jan;14(1):28. PMID: 24703753; PMCID: PMC4104022.*

Figure 4: Cell based-assay with transfected HEK cells with IgLON5 plasmid. A) In red, positive reactivity of a serum of a patient containing IgLON5 antibodies that co-localize with B) green fluorescent protein (GFP) tag indicating the transfected cells and images are merged in C).

Figure 5: Biochemical characterization of pTau in anti-IgLON5 brains. Immunoblot of insoluble tau aggregates extracted from (1) Alzheimer disease prefrontal cortex tissue, (2) hippocampus of the patient with anti-IgLON5 without brainstem pathology, and (3) brainstem of a previously described patient with anti-IgLON5 with the characteristic brainstem tauopathy. Note that the pattern of bands obtained with anti-phospho-tau antibodies clearly differentiates the 2 IgLON5 cases. In the patient with the brainstem pathology, the 74-KDa band is more intense and appears a differential band around 56 KDa that immunoreacts with the 4R antibody. Both IgLON5 cases have 3R and 4R pTau isoforms. pTau = phosphorylated tau. *Taken from Erro ME, Sabater L,*

Martínez L, Herrera M, Ostolaza A, García de Gurtubay I, Tuñón T, Graus F, Gelpi E. Anti-IGLON5 disease: A new case without neuropathologic evidence of brainstem tauopathy. Neurol Neuroimmunol Neuroinflamm. 2019 Dec 11;7(2):e651. doi: 10.1212/NXI.0000000000000651. PMID: 31826985; PMCID: PMC7007636.

Figure 6: **Heat map of affected areas by tau pathology in human brains with anti-IgLON5 disease.** Note the rostro-caudal gradient of severity and the atypical hypothalamic-brainstem tauopathy described in anti-IgLON5 patients. Coronal sections through the amygdala and the lateral geniculate body (a), midbrain (b), pons (c), medulla oblongata [level of olivary nucleus (d) and decussation of pyramids (e)], and cervical spinal cord (f). Scoring of the frequency of tau pathology in sections stained with tau AT8 is based on the number of tangles and threads: red many; orange moderate; orange dots moderate/few; green dots few; and blue dots isolated. A nucleus ambiguus, AC anterior commissure, Amy amygdala, CA anterior horn, CC crus cerebri, CI inferior colliculus, CN cuneate nucleus, CP posterior horn, ER entorhinal cortex, F fornix, GP pallidum, GN gracile nucleus, HC hippocampus, Hyp hypothalamus, LC locus coeruleus, LTN laterodorsal tegmental nucleus, NPB parabrachial nuclei, NS solitary nucleus, ON olivary nucleus, P putamen, PAG periaquaeductal gray, PI pars intermedia, PPN pedunculopontine nucleus, RU nucleus ruber, R raphe nucleus, RF reticular formation, RF(Gi) gigantocellular reticular nucleus, SG substantia gelatinosa, SI substantia innominata, SN substantia nigra, SNT nucleus of spinal tract of trigeminal nerve, STN subthalamic nucleus, TS solitary tract, V trigeminal nucleus, VIII inf inferior vestibularis nucleus, VIII med medial vestibularis nucleus, X dorsal nucleus vagal nerve, XI spinalis spinal accessory nucleus, XII hypoglossal nucleus, ZI zona incerta. *Taken from Gelpi E, Höftberger R, Graus F, Ling H, Holton JL, Dawson T, Popovic M, Pretnar-Oblak J, Högl B, Schmutzhard E, Poewe W, Ricken G, Santamaria J, Dalmau J, Budka H, Revesz T, Kovacs GG. Neuropathological criteria of anti-IgLON5-related tauopathy. Acta Neuropathol. 2016 Oct;132(4):531-43. doi: 10.1007/s00401-016-1591-8. Epub 2016 Jun 29. PMID: 27358064; PMCID: PMC5023728.*

Figure 7: Cellular inflammation in anti-IgLON5 disease. Cellular inflammation was mild to moderate and mainly composed of perivascular and parenchymal CD3 (a) and CD8 positive T cells (b) and few perivascular CD79a positive B cells/plasma cells (c). Parenchymal CD8 T cells were granzyme B positive and granules showed a polarization towards neurons (d; arrows). In addition, neurons showed an upregulation of MHC class I in the reticular formation and olivary nuclei (e, rectangle enlarged in f). Marked microglia activation was found in the HLA-DR staining in tegmentum of medulla oblongata and nucleus olivaris (g), including formation of microglial nodules (h; rectangle in g enlarged in h). Scale bars: 50 µm. *Taken from Gaig C, Sabater L. New knowledge on anti-IgLON5 disease. Curr Opin Neurol. 2024 Jun 1;37(3):316-321. doi: 10.1097/WCO.0000000000001271. Epub 2024 Apr 1. PMID: 38563128; PMCID: PMC11064895. And adapted from* Berger-*Sieczkowski E, Endmayr V, Haider C, Ricken G, Jauk P, Macher S, Pirker W, Högl B, Heidbreder A, Schnider P, Bradley-*

Zechmeister E, Mariotto S, Koneczny I, Reinecke R, Kasprian G, Weber C, Bergmann M, Milenkovic I, Berger T, Gaig C, Sabater L, Graus F, Gelpi E, Höftberger R. Analysis of inflammatory markers and tau deposits in an autopsy series of nine patients with anti-IgLON5 disease. Acta Neuropathol. 2023 Oct;146(4):631-645. doi: 10.1007/s00401-023-02625-6. Epub 2023 Aug 30. PMID: 37646790; PMCID: PMC10499680.

Figure 8: IgLON5-IgG produces irreversible decrease of IgLON5 clusters on the cell surface and cytoskeletal lesions in the cytoplasmic compartment. Rat hippocampal neurons were treated for 3 weeks with control IgG or Patient IgG. IgLON5 clusters of the neuronal surface were labelled with serum from a patient and visualized in green fluorescence (upper row). Note that patient IgG containing IgLON5 antibodies decreased the number of clusters from the cell surface. After permeabilization, staining of neurofilaments evidenced that patient IgG produces cytoskeletal lesions which could represent the first event pathological event leading to neurodegeneration (middle row). Magnification of a dendrite stained with anti-neurofilament antibodies (lower row). On the right, graphical representation (box plot and whiskers) of the quantification of the lesions produced after the incubation of IgLON5-IgG and control-IgG (NHS, normal human serum). Hypoxic conditions applied to neurons during two days produced similar numbers of lesions than IgLON5-IgG.

Figure 9: IgLON5 is shed to the medium and interacts with the other members of the IgLON family. IgLON5 antibodies block these interactions in vitro independently of the percentage content of IgLON5-IgG1 or IgG4 subclasses. Red-labelled soluble IgLON5 incubated in the medium was able to bind to the surface of transfected HEK cells expressing IgLON1, IgLON2, IgLON3, IgLON4 or IgLON5 (A) but not to untransfected HEK cells or when a soluble red-labelled irrelevant protein was incubated (B). Soluble IgLON5 labelled in green binds to the surface of rat hippocampal neurons when incubated in the medium (C) but not an irrelevant protein (D). Preincubation of soluble IgLON5 with IgLON5 antibodies blocked the interaction of IgLON5 with its binding partners. This effect was independent of the IgLON5-IgG subclass predominance (E). Preincubation of soluble IgLON5 with anti-NMDAR antibodies or with the CSF of a patient with Alzheimer's disease did not produce the observed blocking of interactions (F).

Authorship

All authors declare their contribution in the research and article preparation. All authors approved the final article.

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Table 1. Anti-IgLON5 disease composite score (ICS)

Domain	Points*
Bulbar	Partial Score: 0-21
Stridor	0-1-2-6
Central hypoventilation	0-1-2-6
Dysphagia	0-1-2-6
Dysarthria	0-1-2-3
Sleep	Partial score: 0-12
Abnormal movements/behaviors-vocalizations	0-1-2-3
Insomnia	0-1-2-3
Excessive daytime sleepiness	0-1-2-3
Obstructive sleep apnea	0-1-2-3
Movement disorders	Partial score: 0-15
Gait difficulties and falls	0-1-2-6
Chorea	0-1-2-3
Orofacial dyskinesias	0-1-2-3
Other movement disorders. Specify:	0-1-2-3
Cognition	Partial score: 0-12
Cognitive impairment	0-1-2-6
Neuropsychiatric (psychosis, delirium)	0-1-2-6
Other	Partial score: 0-9
Oculomotor abnormalities	0-1-2-3
Dysautonomia	0-1-2-3
Fasciculations	0-1-2-3
Total composite score	0-69

*0: Absent / normal; 1: Mild; 2: Moderate; 3 (or 6): Severe. aThe score was rated 6, instead of 3, when stridor, central hypoventilation, dysphagia, gait difficulties, cognitive impairment or neuropsychiatric manifestations were considered severe, to weight better the more severe disability caused by these symptoms. From https://www.neurology.org/doi/10.1212/WNL.0000000000208101

Figure 1

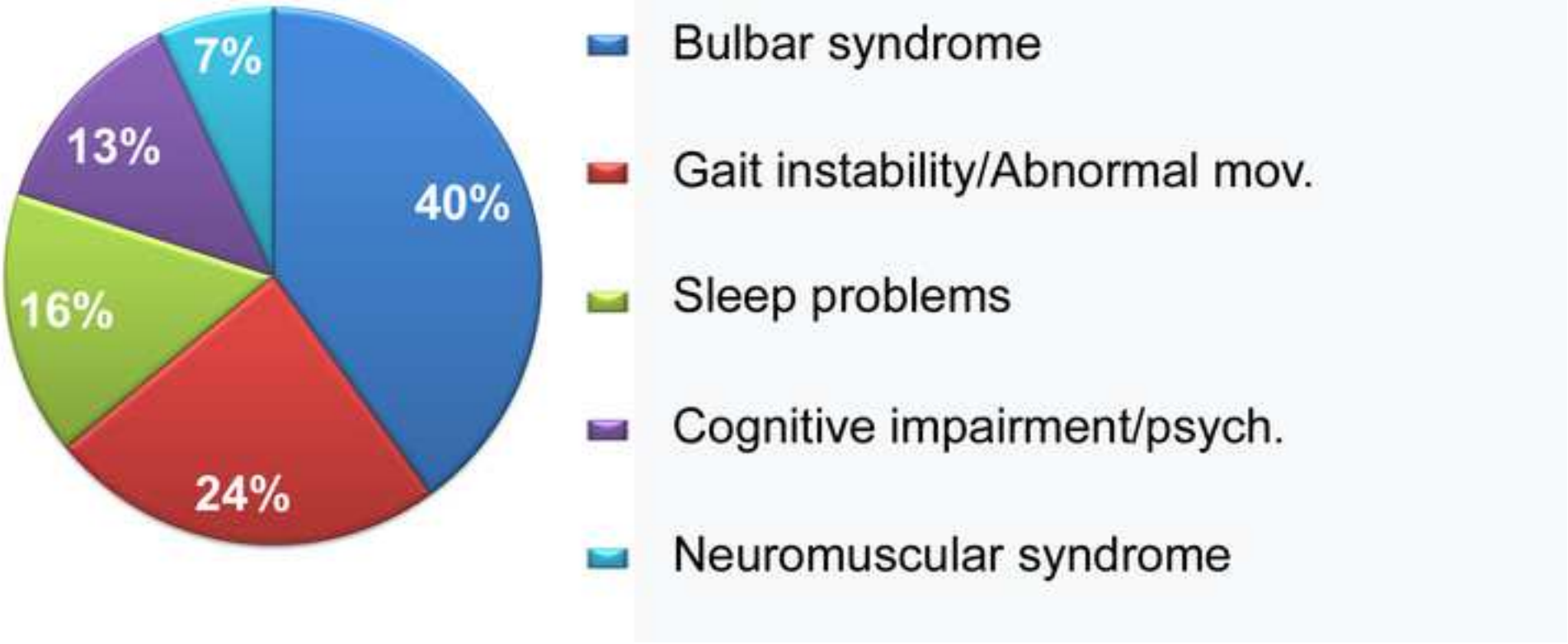

Figure 2

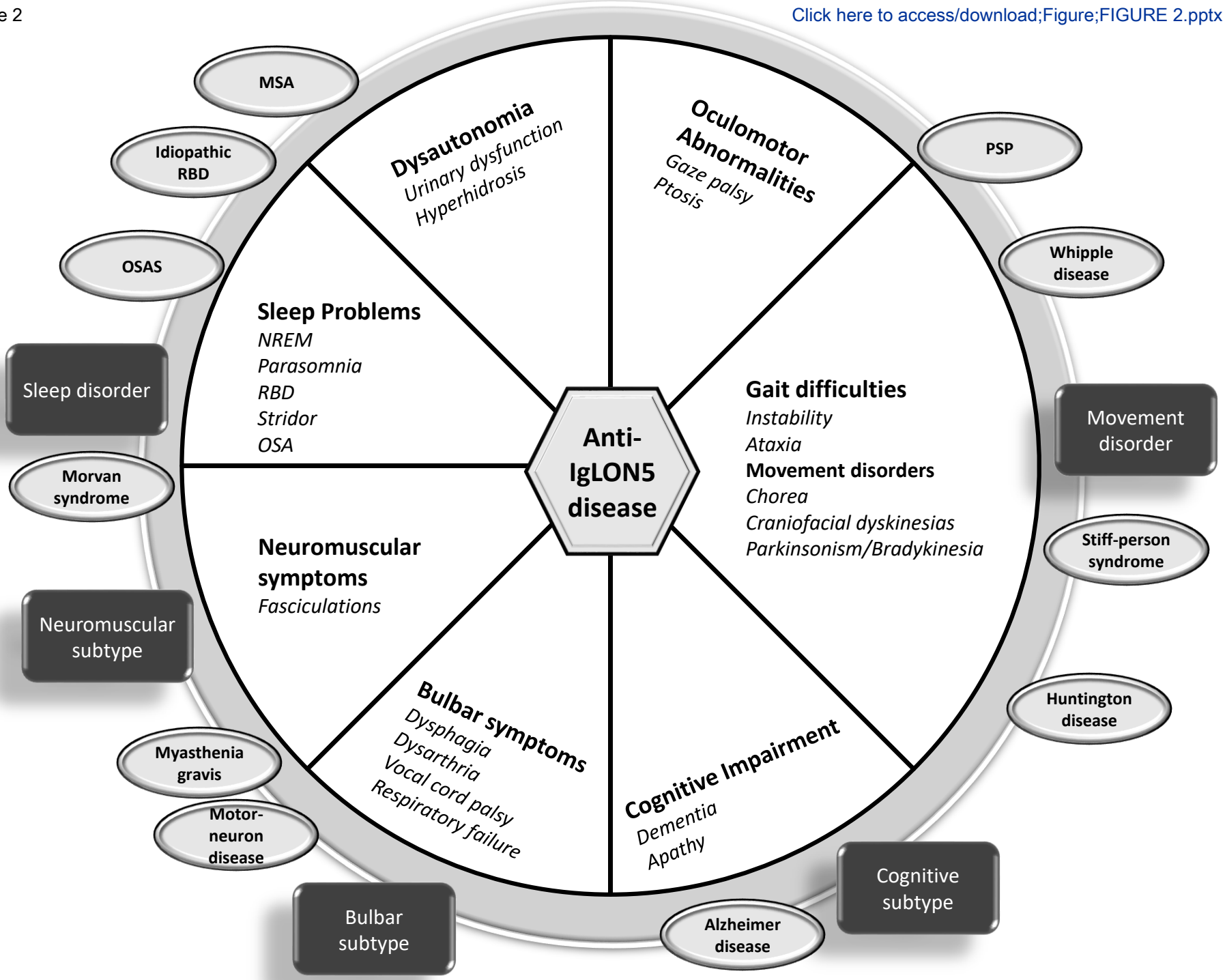

Figure 3

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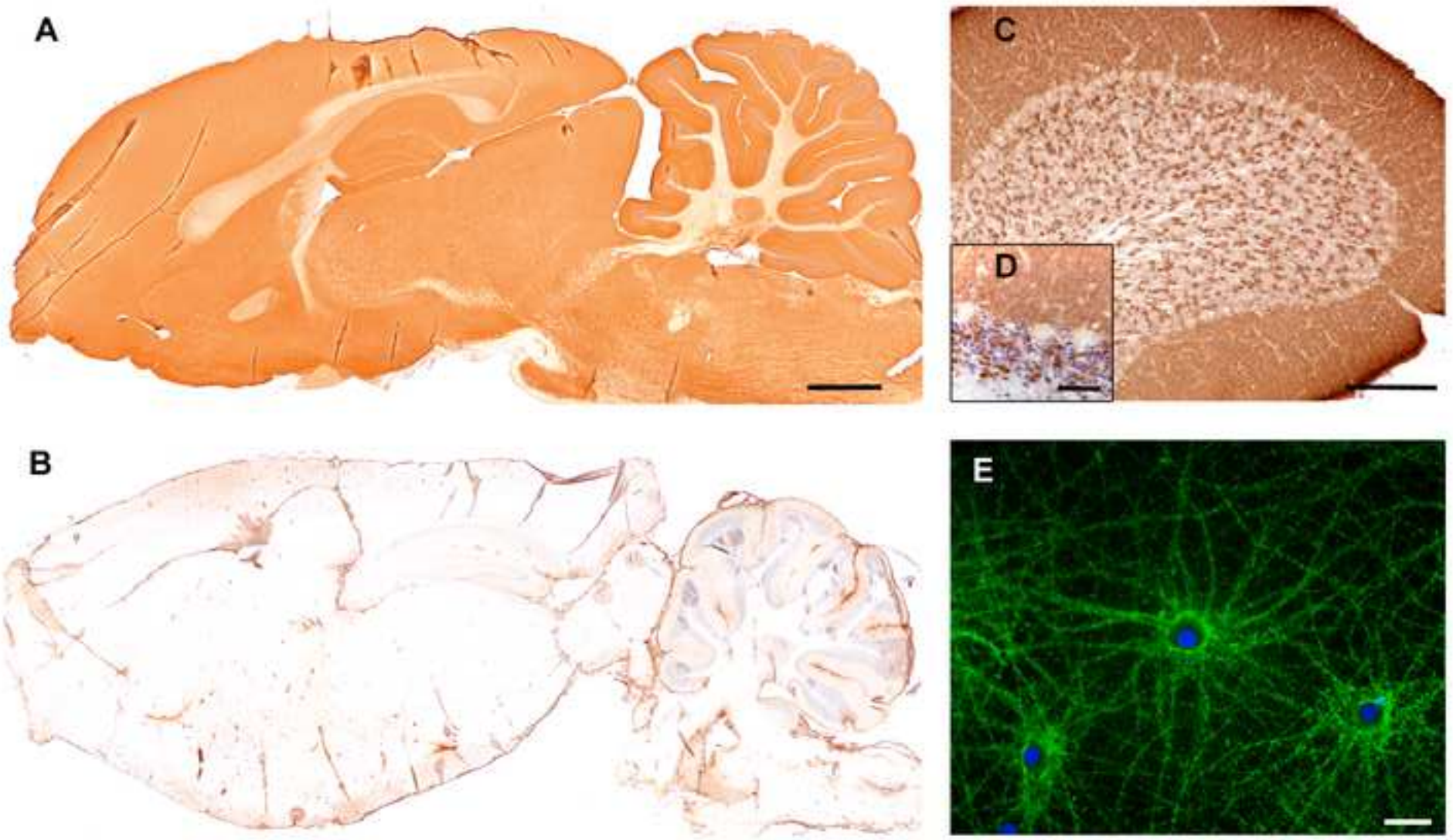

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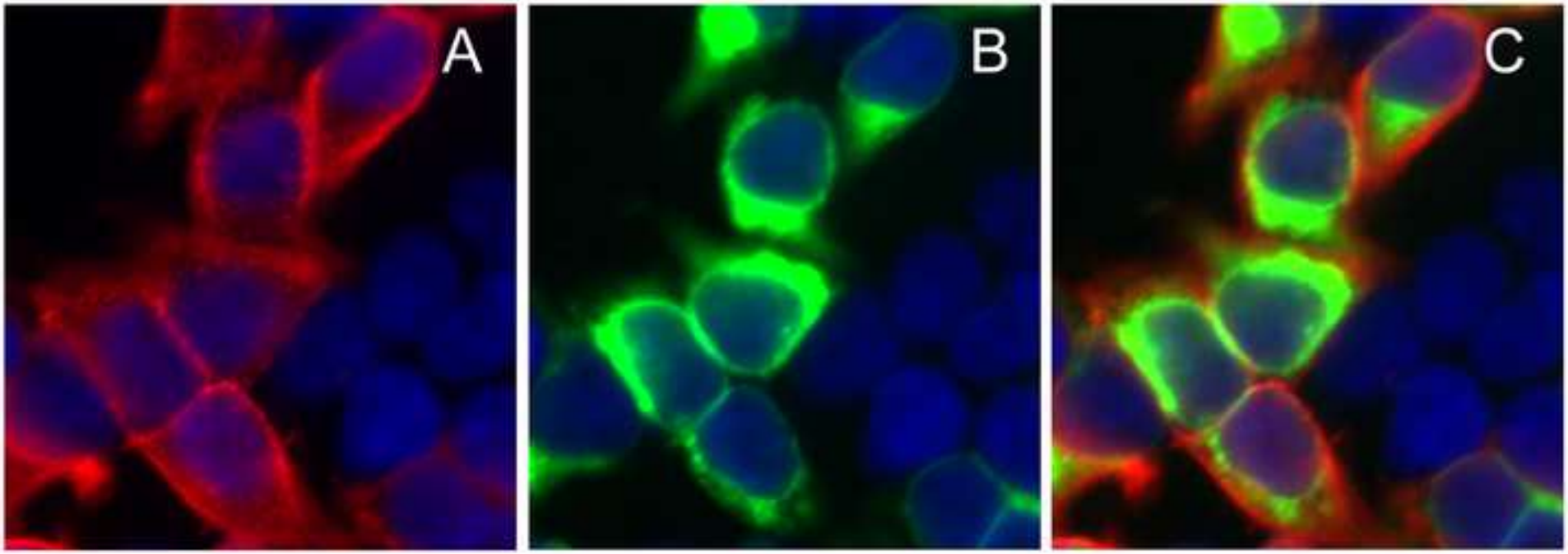

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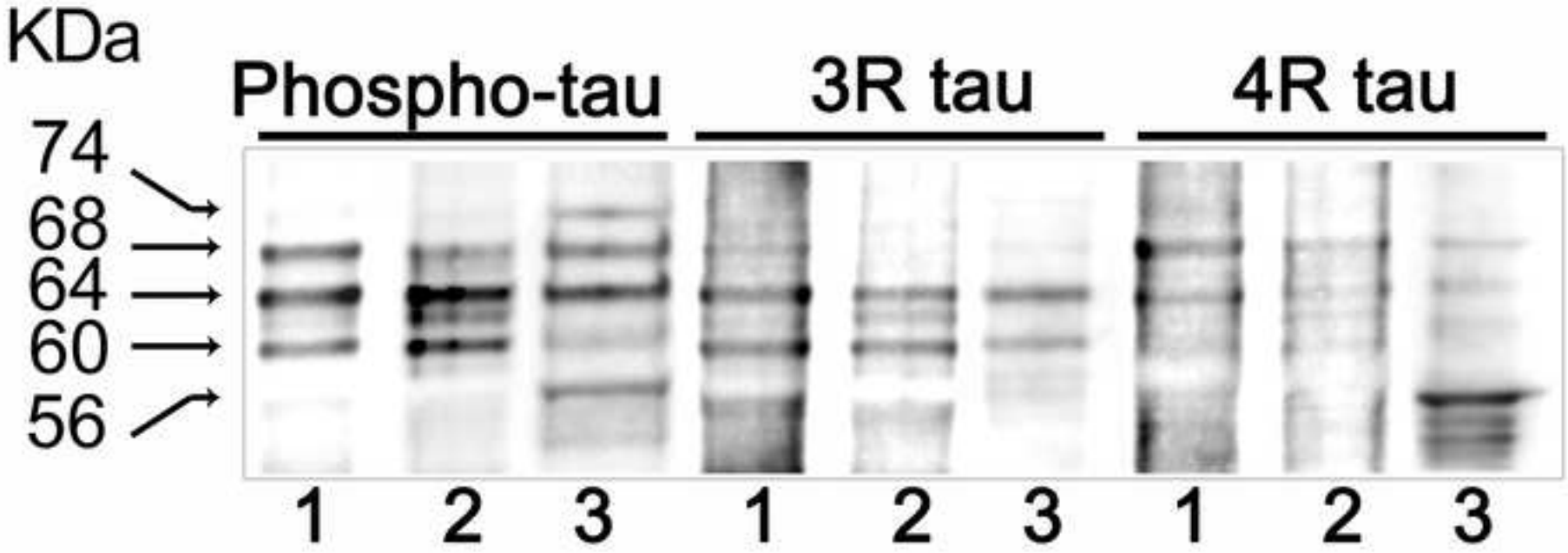

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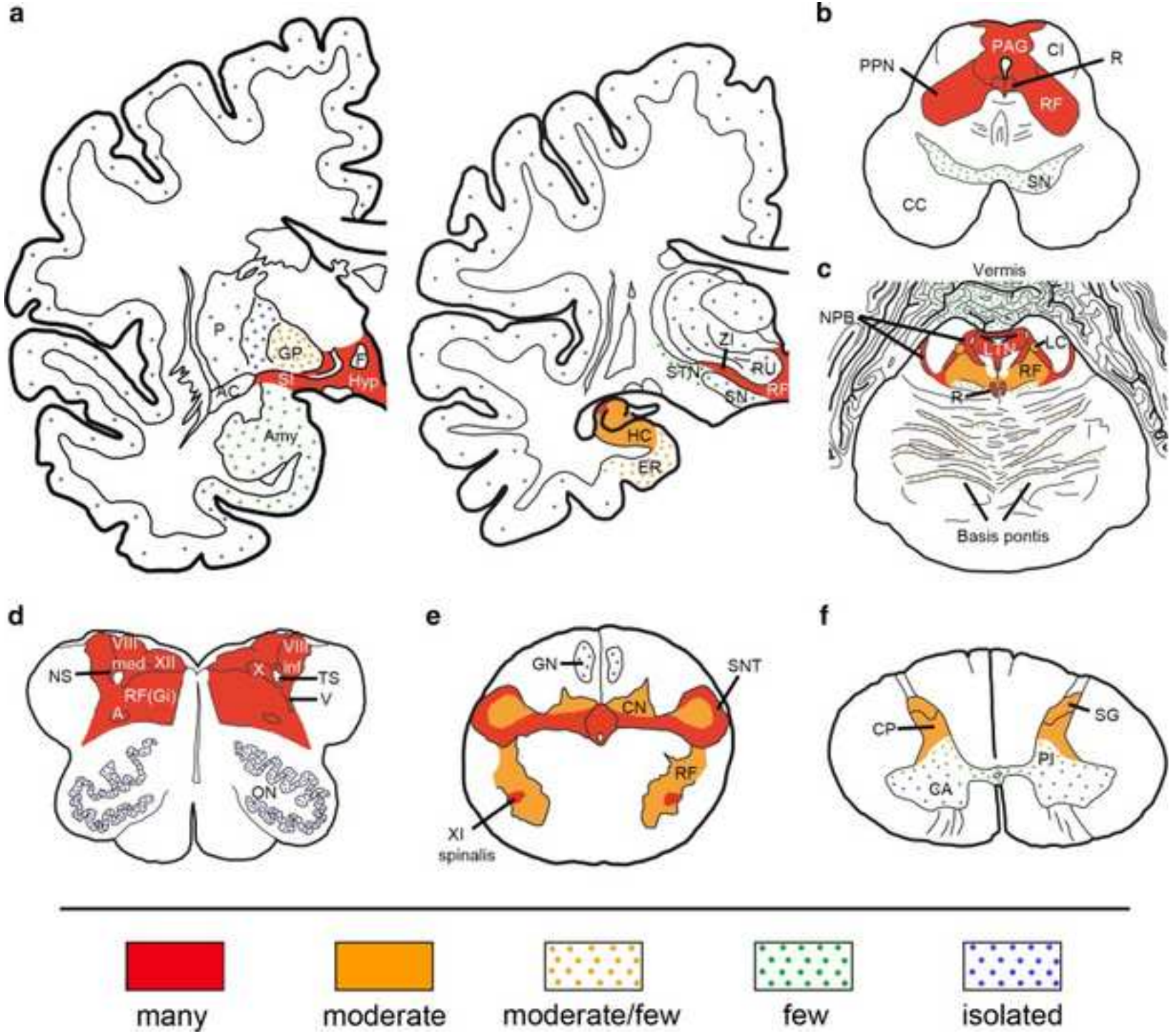

Figure 7

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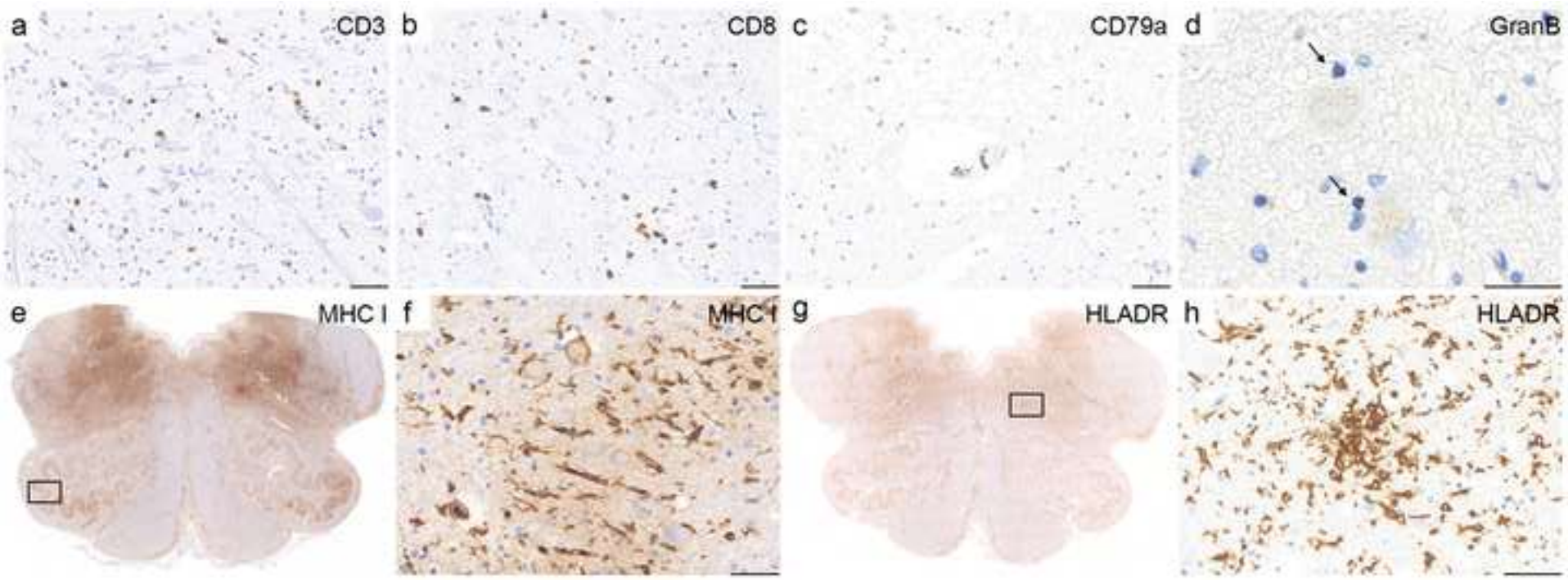

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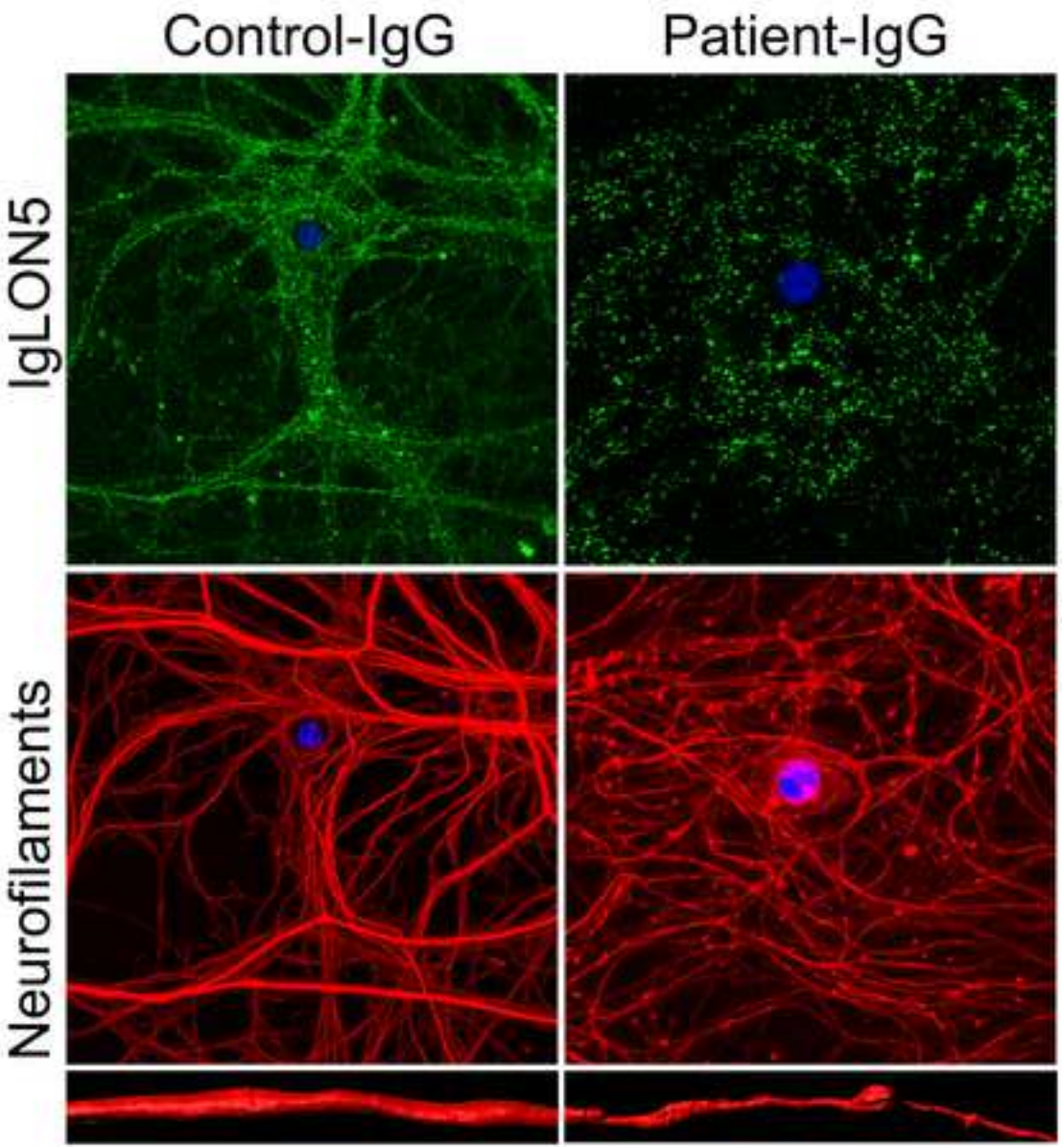

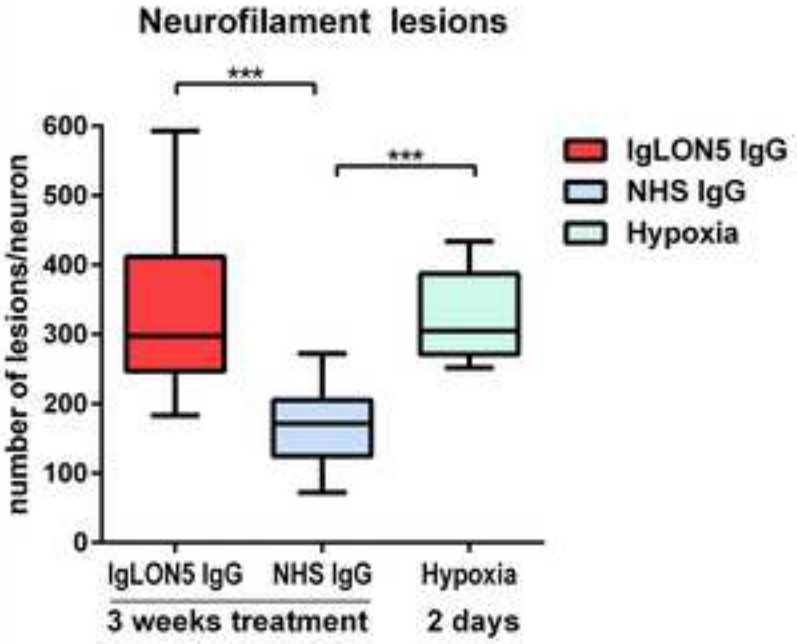

Figure 9

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