MULTIPLE SCLEROSIS JOURNAL | MSJ

Personal Viewpoint

Why does multiple sclerosis only affect human primates?

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Multiple Sclerosis Journal

1–5

DOI: 10.1177/1352458515591862

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Abstract

Background: Multiple sclerosis (MS) develops exclusively in humans. Non-human primates are resistant against MS, although they are highly susceptible to the MS animal model, experimental autoimmune encephalomyelitis (EAE). Unravelling of the cause(s) underlying this discrepancy is highly relevant as insights might be gained into the elusive event(s) that trigger(s) MS. A well-established difference between the human primate (*Homo sapiens*) and non-human primates is that humans are unable to synthesize the sialic acid N-glycolylneuraminic acid (Neu5Gc).

Viewpoint: We propose the concept that long-term ingestion by human primates of the foreign Neu5Gc, via red meat consumption, is an ignored environmental risk factor for MS. Conceptually, incorporation of dietary Neu5Gc into vital regions of the central nervous system, such as the blood–brain barrier (BBB) and the axon–myelin unit, creates targets for binding of de novo synthesized heterophilic anti-NeuGc antibodies. Binding of the antibodies can cause BBB leakage and destabilization of the axon–myelin coupling. The ensuing cytodegeneration and release of self-antigens could be a start of the characteristic pathological features of MS.

Keywords: Axonal loss, demyelination, immunology, progressive, multiple sclerosis

Date received: 5 February 2015; revised: 12 May 2015; accepted: 26 May 2015

Introduction

Although the cause of multiple sclerosis (MS) is not known, it is generally believed that dynamic interactions of genetic and environmental factors, infections in particular, trigger the activation of autoreactive T and B cells present in the normal immune repertoire. A plethora of studies in animal models, the experimental autoimmune encephalomyelitis (EAE) model in particular, shows that autoreactive T and B cells activated in peripheral lymphoid organs can infiltrate the central nervous system (CNS) and trigger cascades of pathophysiological reactions, causing inflammation and tissue injury.[1] However, despite years of intensive research, a single infectious trigger of MS has not been found. This implies that MS may either be induced by a variety of different triggers or, as proposed by us[2] and others,[3] that the cause of MS may lie inside the CNS.

While considering potential triggers of MS, I asked why MS affects only the human primate (HP) and not closely related non-human primates (NHP). To my knowledge, spontaneous development of MS in NHP has not been reported, not even in chimpanzees, despite the >98% genetic similarity with humans. One might argue that in their natural habitat NHP might not be exposed to the environmental triggers of MS. However, even NHP held in captive colonies in moderate climate areas, where they are likely exposed to the same environmental cues as humans, do not develop MS spontaneously. A documented exception may be the spontaneous inflammatory demyelinating disease in a Japanese macaque colony (*Macaca fuscata*) at the Oregon Primate Center.[4] A novel simian γ2-herpesvirus, Japanese macaque rhadinovirus (JMRV), was identified as the potential trigger of the disease. However, definitive proof in deliberately infected monkeys is still lacking.

Are non-human primates resistant against multiple sclerosis?

One could argue that NHP are (genetically) resistant to MS. Our work in the EAE models in marmosets (*Callithrix jacchus*) and rhesus monkeys (*Macaca*

mulatta) over the past 16 years shows no evidence for a low sensitivity of NHP to myelin sensitization; NHP rather seem remarkably sensitive. Marmosets sensitized against the CNS myelin-specific antigen myelin oligodendrocyte glycoprotein (MOG), an immunodominant CNS myelin component, formulated with the relatively weak incomplete Freund's adjuvant, developed an autoimmune neurological disease that shares remarkable clinical and pathological features with MS.[5]

MS risk factors segregate into genetic and environmental factors. To date more than 100 genes with a variable influence on MS have been identified. The vast majority of MS risk genes encode an immunological function, with HLA-DR on top of the hierarchy followed by polymorphisms in the promoter regions of cytokines and chemokines or their receptors.[6] The genetic similarity between HP and NHP is very high, even in highly polymorphic genes, such as those encoding the major histocompatibility complex (MHC) and the antigen receptors of T and B cells. Our studies in common marmosets showed in addition that autoimmunity against myelin involves evolutionarily conserved epitopes. The T-cell epitopes that activate the two dominant pathogenic pathways are MOG24-36, activating MHC class II/ Caja-DRB1*W1201-restricted CD4+ T helper 1 cells, which initiate EAE, and MOG40-48, activating MHC class I/Caja-E restricted cytotoxic T lymphocytes (CTL), which drive EAE progression to irreversible neurological disease.[5] The two epitopes are juxta-positioned in a highly conserved region (residues 20–50) of the MOG extracellular domain. While the pathological relevance of the Th1 pathway in MS is under debate, I noticed that the newly discovered CTL pathway may be implicated in irreversible demyelination by the killing of oligodendrocytes.[7]

Environmental factors enhancing the genetic risk to MS are Epstein Barr Virus (EBV) infection, smoking and possibly vitamin D deficiency.[8] NHP obviously do not smoke, but they express chronic latent infection with EBV-like lymphocryptoviruses (LCV; including EBV in human, CalHV3 in marmosets, macaque LCV in rhesus monkeys). The hairy and pigmented skin of NHP likely limits the natural production of vitamin D from sunlight exposure, even more so in captive colonies housed indoors. Considering these data we regard it rather unlikely that the absence of MS in NHP can be explained by natural resistance.

Are human primates uniquely susceptible to multiple sclerosis?

The NHP EAE model supports the inside-out paradigm, that is, that autoimmunity results from a dysregulated immune reaction against CNS injury; chronic latent infection with cytomegalovirus (CMV) and EBV contribute to the hyperresponsive state.[9] Chronic infection of HP with CMV creates a (progressively expanding) repertoire of highly reactive cytotoxic T lymphocytes with NK cell markers (NK-CTL), which protect against reactivation of the virus.[10] The same holds true for the rhesus monkey. Chronic CMV infection contributes also to changes in the immune system of very elderly people that likely underlie the increasing prevalence of chronic inflammatory diseases.[10] We noticed that the autoreactive CTL identified in the NHP EAE model cross-react with the UL86 ORF-encoded major capsid protein of CMV and that they have a comparable effector memory phenotype and MHC restriction (MHC-E) as the NK-CTL of humans. Marmoset B cells infected with EBV have the capacity to recruit and activate the CTL.[9] These findings indicate that humans may not be uniquely susceptible to MS, as the pathogen-educated immune system of HP and NHP may be similarly responsive to antigen released from a hypothesized primary CNS injury.[3,11]

A human-specific cytodegeneration paradigm

The inside-out paradigm and the notion that MS affects only humans may imply a specifically human degenerative event as etiopathogenic trigger. However, myelin release per se, such as in stroke, usually does not elicit (chronic) autoimmune disease. Recent work shows that elicitation of autoimmunity may be prevented by glycoproteins expressed on the myelin surface (e.g. MOG-Le^x) that bind C-type lectin receptors (e.g. DC-SIGN) of antigen presenting cells (APC) and inhibit APC maturation. The maturation block is removed when the glycan moiety of MOG is changed by inflammatory factors present in early MS lesions.[12]

The vast majority of glycoproteins that decorate body cells terminate by the structurally almost identical sialic acids N-acetylneuraminic acid (Neu5Ac) and N-glycolyneuraminic acid (Neu5Gc)[13] (Figure 1). Neu5Ac is the precursor of Neu5Gc and conversion is catalysed by CMP-N-acetylneuraminic acid hydroxylase (CMAH). Neu5Gc is absent in the CNS as expression of CMAH is suppressed, indicating that expression of Neu5Gc within the CNS may be detrimental.[14] Indeed, overexpression of CMAH in the mouse brain may have detrimental consequences.[15] This warrants the question whether Neu5Gc might be implicated in the destabilization of the axon–myelin unit. Compact myelination of CNS axons is mediated by the interaction of myelin-associated glycoprotein (MAG) with

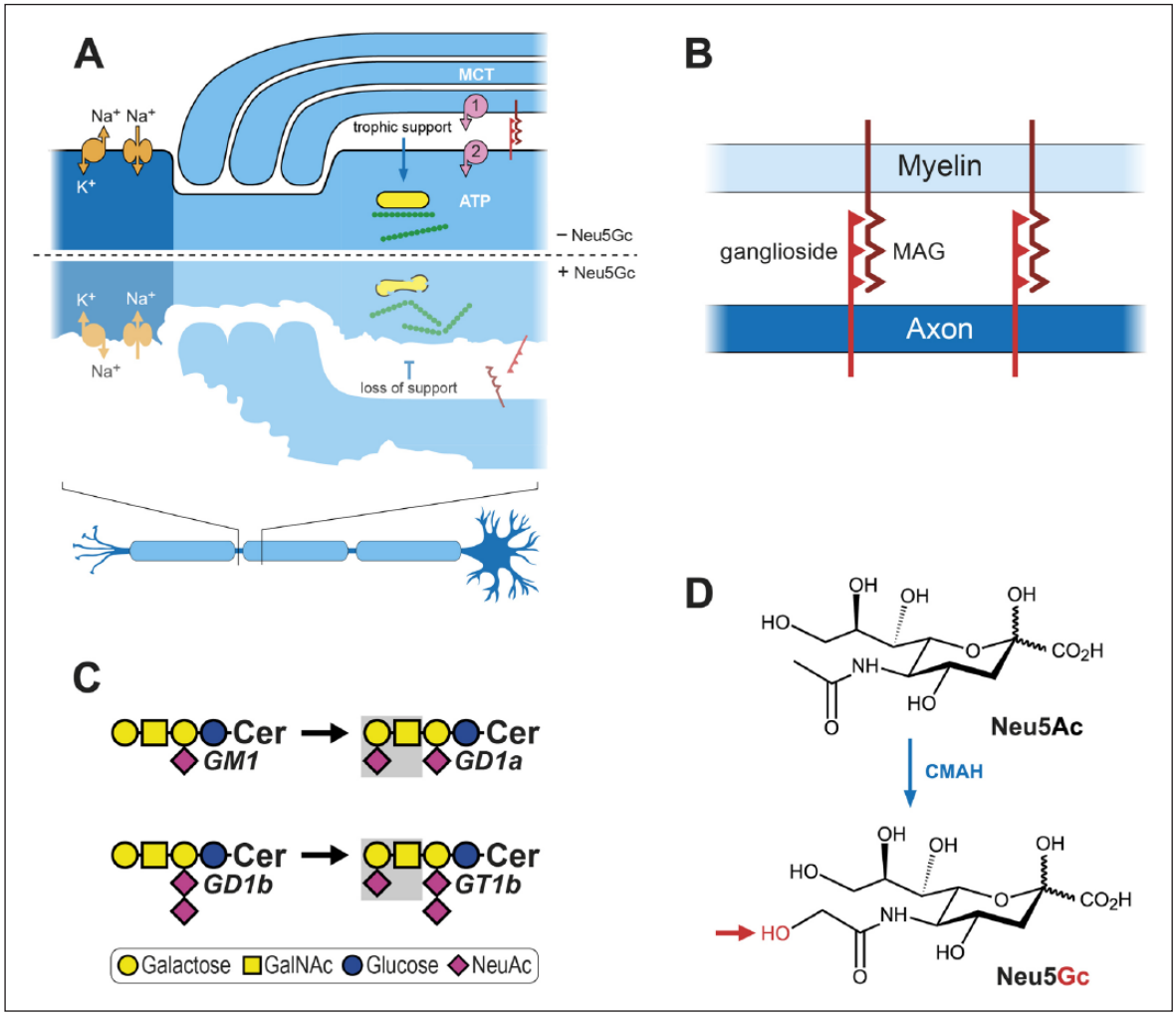

Figure 1. Neu5Gc-induced degeneration of the axon–myelin unit.
Depicted in panel **A** is a graphic representation of the effect that Neu5Gc incorporation may have on the axon–myelin unit; the figure is inspired by Simons et al.[23] **B.** The axon–myelin unit is stabilized by myelin-associated glycoprotein (MAG) binding of gangliosides. MAG is a sialic acid-binding lectin (Siglec-4), with high specificity for N-acetylneuraminic acid (Neu5Ac). Panel **C** depicts the most important gangliosides of CNS axons, which all terminate with Neu5Gc. **D.** The conversion of Neu5Ac into N-glycolylneuraminic acid (Neu5Gc) is catalysed by CMP-N-acetylneuraminic acid hydroxylase (CMAH) and involves substitution of a -H for an -OH group (arrow). Panel **A** depicts the putative effect of Neu5Gc on the axon–myelin unit. The healthy unit (upper half) is stabilized by MAG molecules binding gangliosides expressed on the axon surface. Trophic support is provided from the oligodendrocyte to the axon via transport of monocarboxylates (pyruvate, lactate) through transporters (MCT) in the inner myelin lamellae and the axon. The nutrients are fed into the mitochondria to generate sufficient ATP for satisfying the high energy need of the axon, i.e. electron pulse conduction and vesicle transport to the synapses. The lower half shows the situation when MAG–ganglioside binding is blocked by Neu5Gc incorporation and heterophilic antibody binding. The ensuing dissociation of myelin from the axon antibody impairs trophic support, which causes metabolic problems to the axon. The incapacity to produce sufficient ATP causes a pulse-conduction block, mitochondrial degeneration and problems with Ca^{2+} homeostasis, which inevitably leads to the degeneration of the neuron/axon complex.

gangliosides (GM1, GD1a, GD1b, GT1b) on the axon (Figure 1). MAG is a sialic acid-binding lectin (Siglec4) with high specificity for the Neu5Ac residue that caps the major axonal gangliosides.

Humans are unique among hominoid primates (bonobos, chimpanzees, gorillas, orangutans, gibbons) because of their inability to synthesize Neu5Gc in any of their body tissues due to an irreversible exon deletion in the CMAH gene, which occurred after the most recent branching of hominoid NHP (around 3 million years ago)[14]. Nevertheless, detailed analysis of human necropsy tissues showed presence of Neu5Gc in a variety of healthy and malignant tissues.[16] The incorporated Neu5Gc was shown to come from the consumption of high Neu5Gc-containing food, red meat from livestock (beef, pork and lamb) in particular. A second important finding has been that due to this defect humans generate

antibodies against the xeno-sialic acid (e.g. Hanganutziu-Deicher antibodies in cancer or Paul-Bunnell antibodies in infectious mononucleosis), which are likely induced by gut flora elements incorporating dietary Neu5Gc.[17,18] These heterophilic antibodies were found to bind tissues with incorporated Neu5Gc and elicit low-burning inflammation.[19]

What is the relevance of this paradigm for MS? There is a remarkable overlap of global areas where the MS risk is high with areas where meat from livestock is part of the daily diet. Note also that the dramatic increase of MS incidence in Japan after World War 2 has been attributed to the introduction of the Western diet.[20] While the Neu5Gc content of traditional Japanese food, based on fish, poultry and plants is low, it has been calculated that consumption of 250 g red meat per day equals to daily intake of 4.5–7.5 grams Neu5Gc.[16]

One pathogenically relevant site of Neu5Gc incorporation in the CNS is the blood–brain barrier, where binding anti-Neu5Gc antibody binding may induce endothelial leakage and antibody diffusion into the CNS parenchyma.[21] The second relevant site of Neu5Gc incorporation is the interaction of MAG with axonal gangliosides (GM1, GD1a, GD1b, GT1b). It has been shown in neuroblastoma cell cultures that metabolic substitution of Neu5Ac by Neu5Gc abrogates MAG binding.[22] It remains to be established whether a sufficient proportion of dietary Neu5Gc will be incorporated in the CNS, but heterophilic antibodies binding low numbers of Neu5Gc epitopes will most likely hinder MAG–ganglioside interaction already at low Neu5Gc incorporation.

There is a relatively large body of literature about the consequences when the axon–myelin unit is disturbed, reviewed recently by Simons et al.[23] The organization of the unit as bands of compactly wound myelin interspaced by (non-myelinated) nodes of Ranvier allows the fast saltatory conduction of action potentials, but the price paid is that axons depend on the oligodendrocyte for trophic support. Indeed, astrocytes and oligodendrocytes satisfy the high energy need of axons via the supply of monocarboxylate nutrients (pyruvate, lactate) through MCT transporters in the internodes and axons. The strong dependence of axons on glia for the functioning axon–myelin unit implies that disturbed couplings within the unit may result in its degeneration (Figure 1).

Perspective for validation of the concept

I propose that daily consumption of meat from livestock should be regarded as an environmental risk factor for MS. A great uncertainty in the concept, however, is whether the proportion of orally fed Neu5Gc that incorporates in the brain is sufficient for destabilizing the axon–myelin unit, for the induction of heterophilic antibodies and for activation of an autoimmune process. The common marmoset offers an attractive experimental animal model for answering these questions, as it shares the genetic CMAH defect with humans[24] as well as MS-like autoimmune reactions against human myelin.[25]

Acknowledgements

The author thanks Mr Henk van Westbroek (BPRC) for the artwork and Prof Dr Jeroen J Geurts (VUmc, Amsterdam) for valuable discussions and critical reading of the manuscript.

Conflict of interest

None declared.

Funding

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

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