RELAPSING-REMITTING MULTIPLE SCLEROSIS: ADVANCES IN DISEASE-MODIFYING THERAPIES

by

KATHLEEN KAY

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Approved by

First Reader

John W. Hare, M.D.
Voluntary Faculty
Boston University Medical School

Second Reader

Anne Cross, M.D.
Professor of Neurology
Washington University
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ABSTRACT

Multiple sclerosis is a demyelinating disease affecting the central nervous system. It is the most prevalent disabling neurological condition among young adults, with onset typically between 20 and 40 years of age. Infiltrating immune cells and microglia activations are associated with inflammatory and neurodegenerative mechanisms. Current available disease modifying therapies suppress or modulate the immune system. These pharmaceuticals differ with respect to administration route and frequency, adverse effects, and efficacy. This paper provides a thorough manuscript illustrating the major prescribing factors, efficacy profiles, adverse events, and contraindications that patients and clinicians should consider while choosing a treatment. Despite the advancements made over the past 20 years, patients with progressive multiple sclerosis have few therapeutic options. Additionally, this paper assesses emerging therapies and disease targets on the pharmaceutical horizon, which have shown promise for all disease phenotypes.
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LIST OF ABBREVIATIONS

APCs ................................................................. Antigen Presenting Cells
ARR ................................................................. Annualized Relapse Rate
b.i.d. ................................................................. Twice Daily
CIS ................................................................. Clinical Isolated Syndrome
CNS ................................................................. Central Nervous System
COX-2 ............................................................. Cyclooxygenase-2
CSF ................................................................. Cerebrospinal Fluid
DMF ................................................................. Dimethyl Fumarate
DMT ................................................................. Disease-Modifying Therapy
EAE ................................................................. Experimental Autoimmune Encephalomyelitis
EDSS ............................................................... Expanded Disability Status Scale
ENV ................................................................. Envelope Protein
FDA ................................................................. Food and Drug Administration
FGF-2 ............................................................. Fibroblast Growth Factor 2
FN ................................................................. Fibronectin
GA ................................................................. Glatiramer Acetate
Gd+ ................................................................. Gadolinium-enhancing
GSH ................................................................. Glutathione
HERV-W ......................................................... Human Endogenous Retrovirus Type W
IFNβ ............................................................... Interferon beta
Igs ................................................................. Immunoglobulins
<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Full Form</th>
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<tbody>
<tr>
<td>IL</td>
<td>Interleukin</td>
</tr>
<tr>
<td>iNOS</td>
<td>Nitric Oxide Synthase</td>
</tr>
<tr>
<td>JC</td>
<td>John Cunningham</td>
</tr>
<tr>
<td>LINGO-1</td>
<td>Leucine-rich repeat &amp; immunoglobin domain-containing Nogo receptor-interacting protein</td>
</tr>
<tr>
<td>MAdCAM</td>
<td>Mucosal Addressin Cell Adhesion Molecule</td>
</tr>
<tr>
<td>MBP</td>
<td>Myelin Basic Protein</td>
</tr>
<tr>
<td>MCP-1</td>
<td>Monocyte Chemotactic Protein-1</td>
</tr>
<tr>
<td>MMF</td>
<td>Monomethyl Fumarate</td>
</tr>
<tr>
<td>MRI</td>
<td>Magnetic Resonance Imaging</td>
</tr>
<tr>
<td>MS</td>
<td>Multiple Sclerosis</td>
</tr>
<tr>
<td>NF-κB</td>
<td>Nuclear Factor-κB</td>
</tr>
<tr>
<td>NICD</td>
<td>Notch Intracellular Domain</td>
</tr>
<tr>
<td>Nrf2</td>
<td>Nuclear factor erythroid-derived 2-like 2</td>
</tr>
<tr>
<td>OPCs</td>
<td>Oligodendrocyte Progenitor Cells</td>
</tr>
<tr>
<td>OPN</td>
<td>Osteopontin</td>
</tr>
<tr>
<td>PDGF</td>
<td>Platelet-derived Growth Factor</td>
</tr>
<tr>
<td>PLP</td>
<td>Proteolipid Protein</td>
</tr>
<tr>
<td>PML</td>
<td>Progressive Multifocal Leukoencephalopathy</td>
</tr>
<tr>
<td>PPMS</td>
<td>Primary Progressive Multiple Sclerosis</td>
</tr>
<tr>
<td>RRMS</td>
<td>Relapsing-Remitting Multiple Sclerosis</td>
</tr>
<tr>
<td>ROS</td>
<td>Reactive Oxygen Species</td>
</tr>
</tbody>
</table>
SIP ................................................................. Sphingosine-1-phosphate
SPMS ............................................................ Secondary Progressive Multiple Sclerosis
TGF-β1 ............................................................ Transforming Growth Factor β1
Th ................................................................. T-helper
TLR4 ............................................................. Toll-like Receptor 4
TNF-α ............................................................. Tumor Necrosis Factor α
Treg .............................................................. Regulatory T
VCAM ........................................................... Vascular Cell Adhesion Molecule
I. Introduction

Multiple sclerosis (MS) is a heterogeneous inflammatory disease characterized by plaque formation and the demyelination of the central nervous system (CNS) (Mallucci et al., 2015). Those afflicted experience varying degrees of sensory, motor, and cognitive deficits. According to the National Multiple Sclerosis Society, it is estimated that 400,000 individuals in the United States and 2.3 million individuals worldwide suffer from MS (“MS Prevalence,” n.d.). Believed to be influenced by both genes and the environment, disease onset generally occurs between 20 and 40 years of age, and women have a higher disease prevalence (Kalman and Lublin, 1999). Data suggest that there is a greater prevalence of MS further from the equator, although certain ethnic populations have significantly lower incidences of the disease in spite of their residential latitude (Kalman and Lublin, 1999). Other potential triggers include viral and bacterial infections (Agrawal and Yong, 2007). The etiology and pathogenesis of MS remains a conundrum, despite extensive research.

Diagnostic criteria

No single definitive test exists for the diagnosis of MS. This diagnosis rests upon clinical evaluation and diagnostic tests utilized to rule out other conditions. Classification of MS is dependent on disease activity and progression (Mallucci et al., 2015). Clinical manifestations of disease activity may be too subtle to detect, but closely monitoring cognitive, visual, and other changes helps to identify it (Lublin et al., 2014). Magnetic resonance imaging (MRI) is the most sensitive diagnostic tool and plays an essential role
in the detection of pathological alterations of the central nervous system (CNS). As stated by Radue et al. (2010):

“Magnetic resonance imaging (MRI) may be used to observe the course of multiple sclerosis (MS) by revealing active inflammation and migration of lymphocytes across the blood-brain barrier (appearing as T1 gadolinium-enhancing [Gd+] lesions), overall disease burden (total T2-hyperintense lesion load), and irreversible central nervous system (CNS) damage (indicated by chronic, persistent T1-hypointense lesions and brain volume loss).”

In addition to MRI measures of disease activity, cerebrospinal fluid (CSF) may be analyzed for abnormalities. However, CFS biological markers do not reliably differentiate between MS disease phenotypes. The CSF usually contains elevated levels of antibodies, oligoclonal bands, and certain proteins associated with the breakdown of myelin (Haines et al., 2015).

Lublin et al. (2014) classifies the disease course by one of three clinical presentations, clinical isolated syndrome (CIS), relapsing-remitting multiple sclerosis (RRMS), or progressive multiple sclerosis, subdivided into primary and secondary categories (Table 1). Although not initially included as a clinical descriptor, CIS is as an early presentation of MS that falls short of the clinical diagnosis (Mallucci et al., 2015). CIS is characterized as an “acute or sub acute onset of monophasic episode suggestive of Multiple Sclerosis” (Mallucci et al., 2015, p. 2). While CIS fails to fulfill MS diagnostic criteria, Lublin et al. (2014) reports that 30% to 70% of patients develop MS, with
prevalence varying with location of CIS episode. Accounting for approximately 85% of all MS cases, relapsing-remitting multiple sclerosis (RRMS) fluctuates between periods of symptomatic relapse and complete or partial remission of neurological dysfunction (Lublin et al., 2014). RRMS affects women more frequently than men (Lublin et al., 2014). Lublin et al. (2014) subcategorizes progressive MS into secondary progressive and primary progressive MS. Seventy-five percent of RRMS cases transition to secondary progressive multiple sclerosis (SPMS) within 15 years of onset. It is characterized by a progressive deterioration in neurological function post relapsing phase (Lublin et al., 2014). Primary progressive multiple sclerosis (PPMS) lacks the acute relapsing course of the disease and, therefore, experiences a steady increase in neurological deficits from the initial onset (Lublin et al., 2014). This progression is assessed yearly by clinical and imaging criteria.
Table 1. Characteristics of MS Phenotypes. Adapted from Mallucci et al., 2015.

<table>
<thead>
<tr>
<th>Clinical Form</th>
<th>Disease Course</th>
</tr>
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</table>
| **Clinical isolated syndrome (CIS)**       | Characterized by acute or sub acute onset of monophasic episode suggestive of MS, that has yet to meet the diagnostic criteria. The episode lasts more than 24h and usually affects optic nerve, brain stem or spinal cord. 
Between 30% and 70% patients with CIS develop MS: In patients with optic neuritis CIS conversion to MS varies between 10% and 85%. In patients with brainstem syndrome CIS conversion to MS varies between 50% and 60%. In patients with spinal cord CIS conversion to MS varies between 40% and 60%. Age of onset between 20 and 45 years Female to male ratio between 2:1 and 5:1. |
| **Relapsing-remitting MS (RR MS)**         | Characterized by relapses over days to weeks, followed by complete or partial remissions over months or years ~85% of cases: 
Age of onset between 20 and 30 years Female to male ratio between 2:1 and 3:1                                                                                                                                 |
| **Progressive MS**                         |                                                                                                                                                                                                                   |
| Secondary Progressive MS (SP MS)           | Characterized by progressive accumulation of disability after initial relapsing course of the disease. ~75% of RR MS cases within 15 years of the initial diagnosis                                                                                                                                 |
| Primary Progressive MS (PP MS)             | Characterized by steady functional worsening from the onset of the disease ~15% of cases: 
Later onset than RR MS (by ~10 years) Female to male ratio: 1:1                                                                                                                                             |
Neuropathology

MS is characterized by axonal demyelination and damage as well as neuronal damage associated with the infiltration of immune cells and microglia activation. Chronic inflammation of the CNS leads to the formation of lesions in the white matter and cortical and subcortical grey matter (Lassmann, 2013). Neuronal degeneration and plaque formation varies between patients, as well as between plaques within a patient. Astrocyte function is also affected. Their disturbance may induce an immune response or may alter the vulnerability of oligodendrocytes or neurons (Brosnan and Raine, 2013). Within active lesions, disruption of perivascular and subpial glia limitans results in the loss of astrocyte polarity (Brosnan and Raine, 2013). Astrocytes form glial scars (Brosnan and Raine, 2013).

Early active plaques are heterogeneous and are characterized by extensive immune cell infiltration (Pierson et al., 2012). “The different lesion types can be delineated on the basis of specific myelin protein loss, presence or absence of infiltrating T cells and macrophages, the accumulation of immunoglobulin and complement (signaling a role of pathogenic autoantibodies), and the presence or absence of severe oligodendrocyte degeneration” (Pierson et al., 2012, p. 206). The majority of lesions are located in the periventricular white matter, cerebellum, brain stem, and optic nerves (Pierson et al., 2012). Patients may also have spinal cord lesions (Pierson et al., 2012). Based on pathological studies as well as some radiology studies, spinal cord lesions are mainly present in SPMS and PPMS patients (Nociti et al., 2005).
Oligodendrocytes produce insulating myelin sheaths around axons of the CNS, which allows saltatory nerve conduction (Bradl and Lassmann, 2010). In the event of myelin or oligodendrocyte damage, axonal support and insulation is compromised and deficits in neuronal function ensue (Mallucci et al., 2015). MS is profoundly heterogeneous. Identifying the mechanisms involved in its pathogenesis is of great importance to the development of effective therapies. Inferences about disease mechanisms are often made through the utilization of an animal model of MS, experimental autoimmune encephalomyelitis (EAE) (Agrawal and Yong, 2007). Although the etiology and pathogenesis have yet to be elucidated, current theories suggest that MS is an autoimmune inflammatory disorder, with infiltrating T cells and activated microglia causing degeneration and demyelination.

T cells are necessary for fighting infectious pathogens, but an inappropriate response is to blame in many autoimmune diseases. Known to regulate the development and function of T-helper (Th) cells and oligodendrocytes, the Notch signaling pathway is the foundation of cell-to-cell communication (Bassil et al., 2013). The Notch signaling pathway is comprised of Notch 1, 2, 3, and 4 transmembrane receptors and their Delta-like 1, 3, and 4 and Jagged 1 and 2 ligands (Bassil et al., 2013). These ligands induce proteolytic cleavage of the receptor, releasing the Notch intracellular domain (NICD) (Bassil et al., 2013). NICD translocates to the nucleus and initiates transcription (Figure 1). Delta-like ligands promote Th1 cell differentiation, while Jagged ligands promote Th2 cells (Bassil et al., 2013). Studies suggest that Th1 and Th17 cells are mainly pathogenic
in human and animal models of MS, while Th2 and regulatory T (Treg) cells are anti-inflammatory (Bassil et al., 2013) (Figure 2).
Figure 1. Diagram of Notch signaling pathway.
Source: Bassil et al., 2014
Figure 2. Illustration of CD4⁺ T cell Differentiation
Based of the cytokine milieu and transcription factors (below the arrows), naïve CD4⁺ T cell can differentiate into various Th cells. Adapted from Bassil et al., 2013.
The CNS stringently regulates the trafficking of immune cells, enabling only limited access (Engelhardt and Ransohoff, 2012). Immune cells extravasate from the vasculature into the CNS via interaction between adhesion molecules and their ligands (Engelhardt and Ransohoff, 2012). Leukocytes express surface α4 integrins that interact with their endothelial-expressed ligands: vascular cell adhesion molecule (VCAM)-1 and mucosal addressin cell adhesion molecule (MAdCAM)-1 (Engelhardt and Ransohoff, 2012). This interaction slows the leukocytes and enables diapedesis across the blood-brain barrier. The invasion of autoaggressive cells triggers inflammatory events that recruit additional immune cells, exacerbating the inflammation and damage to the CNS (Engelhardt and Ransohoff, 2012).

Although the mechanisms involved in brain inflammation and immune-mediated tissue injury have not been fully elucidated, it is widely accepted that inflammation is associated with active peripheral T cells that recognize specific CNS molecules. Normally, microglia and dendritic cells, antigen presenting cells (APCs), recognize pathogen-associated molecular patterns, which generate proinflammatory signals to protect against invading pathogens (Agrawal and Young, 2007). If APCs become reactive against endogenous proteins, they stimulate the production of self-reactive T cells, characteristic of many neurodegenerative diseases (Figure 3).
Figure 3. Immune response in MS. This is a hypothetical view of the immune response in acute multiple sclerosis lesions. Proinflammatory cytokines and antigens induce an immune response in the CNS. T and B cells readied in the peripheral lymphoid tissue, then migrate into the CNS. Microglia and CD4+ T cells produce more inflammatory cytokines, and these cytokines attract other immune cells. 

Source: Hemmer et al., 2002
CD4$^+$ and CD8$^+$ T cells play major roles in the immune response. Additionally, an increase in immunoglobulins (Igs) detected in the CSF suggests that both B cells and Igs have a hand in MS pathogenesis (Agrawal and Yong, 2007). Under normal circumstances, B cells are unable to cross the blood-brain barrier; however inflammation causes it to be “leaky” (Agrawal and Yong, 2007). B cells can act as APCs and recruit T lymphocytes to the parenchyma, and they can secrete myelin-specific antibodies causing tissue damage (Agrawal and Yong, 2007). CD8$^+$ T cell infiltrates are the major constituents of MS lesions (Skulina et al., 2004). A positive correlation exists between axonal damage and the number of CD8$^+$ T cell infiltrates, as well as MRI measures of disease and CD8$^+$ T cells detectable in blood (Skulina et al., 2004).

CD4$^+$ T cells play an equally crucial role in pathogenesis. According to a study performed by Tzartos et al. (2008), T cells, astrocytes, and oligodendrocytes increased the expression of interleukin (IL)-17 in areas of inflammation and active demyelinating lesions. Within acute lesions and along the active borders of chronically active ones, significantly greater densities of both CD4$^+$ IL-17$^+$ and CD8$^+$ IL-17$^+$ T cells were observed (Tzartos et al., 2008). Observed by Zhang et al. (2015), the production of IL-11, along with IL-17, was significantly increased in T lymphocytes in RRMS, suggesting that it too plays a role in the proinflammatory response. Demonstrated in this study, Th17 cytokines IL-17F, IL-21, and tumor necrosis factor α (TNF-α), and transforming growth factor β1 (TGF-β1) induced the differentiation of IL-11$^+$CD4$^+$ T cells. IL-17F, TNF-α, TGF-β1, IL-1β promoted the secretion of IL-11 from CD4$^+$ T cells (Zhang et al., 2015).
TNF-α-induced IκB phosphorylation activates the nuclear factor (NF)-κB pathway, which upregulates the expression of proinflammatory chemokines, such as monocyte chemotactic protein-1 (MCP-1), and adhesion molecules, including VCAM-1 and E-selectin (Wei et al., 2015). All of these molecules are necessary for the attraction, adhesion, and diapedesis of white blood cells out of the vasculature. Cyclooxygenase-2 (COX-2) and nitric oxide synthase (iNOS), major components of the inflammatory response, are likewise target genes of the NF-κB pathway (Wei et al., 2015).

Furthermore, a reduction of molecules involved in water homeostasis, glutamate buffering, and energy coupling with oligodendrocytes may exacerbate demyelination and neuronal damage (Lassmann, 2013). In order to produce the necessary proteins required for myelination, oligodendrocytes have an extremely high metabolic rate. An unavoidable by-product of cellular respiration, excess reactive oxygen and nitric oxide species may lead to detrimental mitochondrial injury. Activated macrophages also produce nitric oxide and glutamic acid (Bruck, 2005). Excessive nitric oxide exposure causes a conduction block in demyelinated axons, axonal degeneration, and damage to axonal membranes, which may cause ruptures (Bruck, 2005). Axonal damage occurs early in lesion formation and mainly in areas of acute inflammation and demyelination (Bruck, 2005; Ferguson et al., 1997).

**Disease-modifying therapies**

Currently, the main objective of MS disease-modifying therapies (DMTs) is to retard the progression of neurodegeneration by modulating or suppressing the autoimmune response. The approved, available DMTs have only elicited proven
beneficial results in RRMS patients (Wingerchuk and Carter, 2014). With few options for the progressive phenotypes, current research efforts are directed towards compounds with regenerative and neuroprotective properties. These immunomodulatory and immunosuppressant drugs can partially ameliorate disease outcome, but they are in no way a cure and are associated with a plethora of side effects and risks. Patients and their clinicians must weigh the costs and benefits of each treatment option and decide on the one most suited to the patient’s lifestyle.

Goals

Unfortunately, no single pathogenetic mechanism has been identified that would enable the development of a universal therapeutic approach. Studies suggest the MS has a multifactorial etiology. In addition, MS is highly heterogeneous within and between patients, not only in pathogenesis, but also in response to DMTs. This further complicates the treatment process. With considerable published research relating to the safety and efficacy of MS pharmaceuticals and several drugs in various stages of clinical trials, a comprehensive analysis would benefit patients and clinicians alike. The objective of this paper is to provide the reader with a comprehensive manuscript that emphasizes safety, efficacy, and major prescribing factors patients and clinicians should consider while devising a personalized treatment plan. The hope is that they will have a greater understanding of the complex mechanisms that contribute to MS pathogenesis as well as the need for individually tailored approaches for disease management.
II. Clinical Effectiveness of MS Disease Modifying Therapeutics

Approved in the early to mid 90s, self-injectable DMTs were the first available therapies used for the treatment of RRMS (Wingerchuk and Carter, 2014), and even with the recent introduction of oral medications, these injectable DMTs typically serve as the primary treatment for RRMS (Sedal et al., 2014). Due to this twenty-year period of usage, the adverse event profiles and efficacy and safety profiles are well documented, which provides patients and clinicians with a comprehensive therapeutic representation that the newer medications lack (Sedal et al., 2014). Interferon beta (IFNβ) and glatiramer acetate (GA) are widely utilized as first-line therapies for the management of RRMS, but they have relatively low adherence rates and moderate efficacy. Although commonly prescribed, other therapies have been proven more efficacious for curbing clinical and MRI measures of disease progression. The following studies outline the mechanisms of action, effectiveness, safety, and contraindications of the Food and Drug Administration (FDA) approved RRMS medications. Available treatments, their dosing regimes, monitoring suggestions, and adverse events are listed below (Table 2).
Table 2. Current available medications for relapsing-remitting multiple sclerosis. Factors to consider when choosing a therapeutic strategy. Full blood count (FBC), liver function tests (LFT), subcutaneous (s.c.), intramuscular (i.m.). Adapted from Sedal et al., 2014.

<table>
<thead>
<tr>
<th>Medications</th>
<th>Dosing</th>
<th>Frequency of dosing</th>
<th>Adverse effects</th>
<th>Monitoring</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Injectable medications</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>IFNβ-1b</td>
<td>250 µg</td>
<td>Every other day, s.c.</td>
<td>Injection site reactions, flu-like symptoms, leukopenia, elevated liver enzymes,</td>
<td>FBC and LFT at baseline, 1,3, and 6 months then yearly</td>
</tr>
<tr>
<td>IFNβ-1a</td>
<td>30 µg</td>
<td>Weekly, i.m.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IFNβ-1a</td>
<td>22-44 µg</td>
<td>3x/week, s.c.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>IFNβ-1a</td>
<td>125 µg</td>
<td>Every 2 weeks, s.c.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glatiramer acetate</td>
<td>20 mg</td>
<td>Daily, s.c.</td>
<td>Injection site reactions, transient systemic post-injection reactions</td>
<td>FBC and LFT at baseline and 1 month then yearly</td>
</tr>
<tr>
<td></td>
<td>40 mg</td>
<td>3x/week, s.c.</td>
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<tr>
<td><strong>Intravenous medications</strong></td>
<td></td>
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<tr>
<td>Natalizumab</td>
<td>300 mg</td>
<td>Every 4 weeks</td>
<td>Allergic reactions, fatigue, liver toxicity, headache, fatigue, PML</td>
<td>JCV testing (every 3-6 months if negative), MRI at baseline then yearly (3-6 months if JCV positive), FBC and LFT at baseline, 3 months and then biannually</td>
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<tr>
<td><strong>Oral medications</strong></td>
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<tr>
<td>Fingolimod</td>
<td>0.5 mg</td>
<td>Daily</td>
<td>Bradycardia and conduction block during initiation, elevated liver enzymes, risk</td>
<td>Cardiac monitoring at initiation Eye examination at baseline and 3-6 months</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>of herpes infections, macular edema, PML (rare)</td>
<td>FBC and LFT at baseline, 1 month, and then every 4 months</td>
</tr>
<tr>
<td>Dimethyl fumarate BG12</td>
<td>240 mg</td>
<td>Twice daily</td>
<td>Flushing, gastrointestinal events, PML (rare)</td>
<td>FBC and urine protein at baseline, 1 month then 6-12 months</td>
</tr>
<tr>
<td>Teriflunomide</td>
<td>7-14 mg</td>
<td>Daily</td>
<td>Diarrhea, nausea, headache, nasopharyngitis, fatigue, alopecia, elevated alanine</td>
<td>FBC and LFT monthly for 6 months then every 6-8 weeks</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>aminotransferase levels, pain in arms, legs, and back, and PML (rare)</td>
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</table>
Interferon beta

Four preparations of IFNβ have been approved for the treatment of RRMS (Mallucci et al., 2015; “Plegridy,” n.d.). A 250 µg dose of subcutaneous IFNβ-1b is administered every other day, while subcutaneous IFNβ-1a 22 µg or 44 µg is administered three times a week, subcutaneous IFNβ-1a 125 µg every 2 weeks, and intramuscular IFNβ-1a 30 µg, weekly (Mallucci et al., 2015; “Plegridy,” n.d.).

Interferons are naturally occurring anti-inflammatory cytokines (Kieseier, 2011). Although the mechanism is incompletely understood, IFNβ upregulates the anti-inflammatory cytokine expression and downregulates the pro-inflammatory cytokine expression (Kieseier, 2011). A study by Chen et al. (2009) suggests that, in addition to the modulation of Th1 and Th2 groups, IFNβ has the ability to inhibit the production of osteopontin (OPN), a mediator of leukocyte activation and cytokine production, and IL-17 in CD4⁺ T cells.

In a double-blind placebo controlled study, subcutaneous IFNβ-1a was investigated for its safety and efficacy in the treatment of RRMS (Ebers, 1998). The PRISMS study illustrated that subcutaneous IFNβ-1a reduced disease exacerbations and delayed disability progression over the two-year trial course (Ebers, 1998). Compared to the placebo, IFNβ-1a 22 µg and 44 µg significantly reduced the number of relapses per patient during the 2-year trial period by 27% and 33%, respectively (Ebers, 1998). A reduction in the number of moderate and severe relapses was also greatly reduced with IFNβ-1a treatment (Ebers, 1998). Significant reductions in Expanded Disability Status Scale (EDSS) and increases in sustained progression were observed in both IFNβ-1a
groups (Ebers, 1998). Compared to the placebo group, patients in the low-dose IFNβ-1a group had 67% fewer T2 hyperintense lesions, while the high-dose IFNβ-1a group had 78% fewer lesions (Ebers, 1998).

Headache, influenza-like symptoms, generally mild injection-site reactions, fatigue, myalgia, and fever were the most common adverse events (Ebers, 1998). Asymptomatic decreases in white blood cells, neutrophils, and lymphocytes, and slightly elevated liver enzyme levels were observed (Ebers, 1998).

**Glatiramer acetate**

Twenty milligrams of subcutaneous glatiramer acetate (GA) administered daily is indicated for the treatment of RRMS (Mallucci et al., 2015). It is a random polymer composed of glutamic acid, lysine, alanine, and tyrosine, which are found in myelin basic protein (Mallucci et al., 2015). As of January 2014, the FDA approved a 40 mg dose of GA injected three times per week (“Copaxone,” n.d.).

GA functions as an immunomodulator; however, the mechanism remains incompletely understood (Mallucci et al., 2015). GA treatment elicits protective anti-inflammatory responses in dendritic cells and monocytes. Based on a study by Vieira et al. (2003), GA-treated dendritic cells produced fewer TNF-α inflammatory cytokines. Additionally, GA exhibited an inhibitory effect on monocytes and promoted a shift from Th1 inflammatory cells to Th2 anti-inflammatory cells (Weber et al., 2004). Wei et al. (2015) illustrated that GA significantly attenuates TNF-α induced phosphorylation of IκBα, which inhibits its breakdown and prevents the translocation of NF-κB into the
nucleus and activation of NF-κB induced gene transcription. These findings suggested that GA functions via NF-κB inhibition and chemokine production attenuation (Wei et al., 2015).

During a 2-year placebo-controlled phase III trial, patients treated with GA experienced a significant reduction in mean relapse rate compared to those in the placebo group (Johnson et al., 1995). Although significant differences in the mean number of days until relapse and the proportion of relapse-free patients were not observed between the two treatment groups, highly favorably trends were observed for GA therapy (Johnson et al., 1995). More GA-treated patients showed improvement in their EDSS score, while patients in the placebo group were more likely to experience a progression of one or more points (Johnson et al., 1995).

The most common adverse events were localized injection site reactions, including itching, swelling, and erythema (Johnson et al., 1995). Palpitation, tightness around the chest, dyspnea, and associated anxiety occurred sporadically during or within minutes of an injection, generally persisted between 30 seconds and 30 minutes, and passed with no residual difficulties (Johnson et al., 1995). This unpredictable reaction was the only adverse event to occur significantly more often in the GA-treated patients than in the placebo-treated ones (Johnson et al., 1995). Overall, GA treatment was well tolerated in MS patients.

Both GA and IFNβ significantly reduce the relapse rate and disability progression. They have long been the preferred first-line therapies and, not surprisingly, have undergone several head to head efficacy and safety comparisons. Five trials
(BECOME, BEYOND, REGARD, Calabrese, and COMBIRX) directly compared IFNβ and GA over the course of 2 years, with the exception of the 3-year COMBIRX trial (La Mantia et al., 2014). A review of these randomized controlled trials concluded that, in relation to clinical efficacy and safety, IFNβ does not appear to differ from GA (La Mantia et al., 2014). However, all of these studies observed a greater benefit on MRI parameters of disease using in the IFNB arm. It observed a significant reduction in the number of enlarging T2 hyperintense lesions of IFNβ-treated patients (La Mantia et al., 2014).

**Natalizumab**

A 300 mg dose of intravenous natalizumab is administered every four weeks for the treatment of RRMS (Mallucci et al., 2015). Natalizumab, a humanized monoclonal antibody, selectively inhibits the α4 subunits of α4β1 and α4β7 integrins and prevents their interaction with VCAM-1 and mucosal addressin-cell adhesion molecule 1, respectively (Polman et al., 2006). This DMT stabilizes the blood-brain barrier. The diapedesis of lymphocytes into the brain is facilitated by the binding of the α4β1 integrin, on the surface of lymphocytes, and the VCAM-1, on the surface of vascular endothelial cells (Polman et al., 2006). The adhesion of mononuclear cells to the endothelium requires α4β1 integrins (Yednock et al., 1992). According to Yednock et al., the α4β1 integrin contains binding domains for fibronectin (FN) and VCAM-1 adhesion (Yednock et al., 1992). Although the FN-binding site is not involved in adhesion, FN may facilitate leukocyte migration across the wall of the vasculature (Yednock et al., 1992).
Natalizumab has a rapid onset and has advantageous effects on relapse rate, lesion activity, and progression of disability in RRMS patients. Illustrated in the study by Polman et al. (2006), natalizumab reduced the Annualized Relapse Rate (ARR) by 68% after one year of treatment, which was maintained at the two-year study end point. Sixty-seven percent of patients treated with the intravenous drug were relapse free at two years, which was significantly greater than the 41% of patients in the placebo group (Polman et al., 2006). The monoclonal antibody diminished the risk of disability progression by 42% to 54% (Polman et al., 2006). In comparison to the placebo, natalizumab significantly decreased the amount of new or enlarging T2-hyperintense lesions and the amount of Gd+ lesions over two years (Polman et al., 2006).

Polman outlines the frequencies of adverse events. Occurring in 10% or more of the patients, headache, fatigue, arthralgia, depression, infection, and allergic reaction were the most common adverse events (Polman et al., 2006). Fatigue and allergic reaction occurred with significant frequency in patients treated with natalizumab versus the placebo. However, the rate of infection between the natalizumab and placebo groups was insignificant, with infection occurring 1.52 and 1.42 per patient-year, respectively and serious infections occurring in 3.2% and 2.6% of patients (Polman et al., 2006). Relapse and cholelithiasis were the most common serious adverse events. Similar proportions of patients in both the natalizumab and placebo groups discontinued the study due to adverse events (Polman et al., 2006).

Observed in a study by Bloomgren et al. (2012), progressive multifocal leukoencephalopathy (PML) is associated with use of natalizumab in the presence of
several risk factors. Caused by the John Cunningham (JC) virus, PML is an infection of
the brain that attacks the oligodendrocytes resulting in severe disability and death
(Bloomgren et al., 2012). Positive anti-JC virus status, prior use of immunosuppressants,
and increased duration of natalizumab treatment correlated with an increased prevalence
of PML (Bloomgren et al., 2012). It was estimated that the incidence of PML in patients
with a positive anti-JC virus status was greater by a factor of forty-fold than patients
negative for anti-JC virus antibodies (Bloomgren et al., 2012). Patients with a negative
anti-body status incur essentially no risk; however, viral statuses may change throughout
the course of treatment. PML risk increases with increasing treatment duration, with the
greatest risk occurring in populations on natalizumab therapy over 2 years (Bloomgren et
al., 2012). While any of these three factors incur a significant risk, patients with a
combination of all risk factors have the highest estimated occurrence of PML (Bloomgren
et al., 2012).

With 50-60% of the average population testing positive for anti-JC virus
antibodies and infection increasing with age, natalizumab use in RRMS patients should
be monitored closely (Sedal et al., 2014). According to Sedal et al. (2014), current
practices indicate MRI scanning of anti-JC virus antibody positive patients every 4
months and negative patients every 6 to 12 months with biannual clinical status checks.
Additionally, leukocyte levels should be within the normal range prior to starting
treatment. The presence or absence of anti-JC virus antibodies, prior usage of
immunosuppressive therapies, and the severity of the patient’s MS should be taken into
consideration while determining the suitability of natalizumab therapy (Bloomgren et al.,
If viable candidates, patients with positive antibody statuses treated with natalizumab for longer than two years may want to consider other treatment options.

**Fingolimod**

Taken orally, fingolimod was the first MS DMT of its kind. A 0.5 mg dose is administered daily (Mallucci et al., 2015). Fingolimod inhibits the egress of lymphocytes from lymph nodes, rendering them unavailable to initiate an inflammatory event (Kappos et al., 2010). Fingolimod accomplishes this through sphingosine-1-phosphate (SIP) pathway potentiation (Kappos et al., 2010). According to the FREEDOMS study, fingolimod significantly reduced the ARR 54% at a dose of 0.5 mg at the two-year study end when compared with the placebo (Kappos et al., 2010). Witnessed as early as three months post treatment initiation, both trial doses of fingolimod lessened the risk of disability progression over the course of the study (Kappos et al., 2010). Patients treated with fingolimod had significantly fewer Gd+ lesions, fewer new or enlarging T2 hyperintense lesions, and a reduction in T2 hyperintense lesion volume (Kappos et al., 2010).

The proportion of patients that experienced adverse events was comparable across the study groups. The most common adverse events, occurring 10% or more of patients, included infection, headache, abnormal liver function, fatigue, and back pain (Kappos et al., 2010). Abnormally low levels of white blood cells were observed in patients taking fingolimod, which may lead in an increase in the incidence of infection (Kappos et al., 2010). Seen in the FREEDOMS extension trial, the resurgence of latent herpes virus infections was of special interest (Kappos et al., 2015). Bradycardia, MS relapse,
macular edema, and basal cell carcinoma were the most common serious adverse events, occurring in less than 1% of patients (Kappos et al., 2010). According to the FREEDOMS extension trial, the cardiovascular episodes were generally observed within six hours of the administration of the first dose, but resolved and did not recur (Kappos et al., 2015).

Numerous important contraindications require specific laboratory tests and assessments prior to treatment initiation. A patient’s personal and familial history of cardiovascular disease should be taken into consideration prior to treatment with fingolimod. Patients with a heart block or taking drugs that alter atrioventricular conduction should receive special attention. Fingolimod requires cardiac monitoring via electrocardiography following the administration of the first dose (Sedal et al., 2014). Leukocyte counts should be measured prior and monitored throughout treatment duration. Additionally, it is advisable to test for herpes zoster antibodies, and if the patient has a negative status, vaccination is recommended (Sedal et al., 2014). Liver function should be regularly tested, and patients with hepatic or renal impairment may require dose adjustments. Ophthalmologic and dermatologic evaluations are recommended post initiation of fingolimod therapy (Sedal et al., 2014).

The TRANSFORMS study compared the usage of oral fingolimod with intramuscular IFNβ-1a for the treatment of RRMS (Khatri et al., 2011). In the core study, patients were randomly assigned to one of three groups: 0.5 mg daily fingolimod, 1.25 daily oral fingolimod, or 30 µg weekly intramuscular IFNβ-1a (Khatri et al., 2011). In the 13-24 month extension, patients in the IFNβ-1a group were reassigned to either
fingolimod group, and the patients initially treated with fingolimod continued with their treatment protocol (Khatri et al., 2011). Patients who switched to fingolimod from IFNβ-1a had reductions in ARR of 30% on a 0.5 mg daily dose (Khatri et al., 2011). Significantly fewer patients had relapses post switch than during the 12 months of IFNβ-1a treatment (Khatri et al., 2011). Switching to either fingolimod group resulted in significant reductions in the number of new or enlarging T2 hyperintense lesions (Khatri et al., 2011). Additionally, more patients were free from new or enlarging T2 hyperintense lesions (Khatri et al., 2011). The number of new or enlarging Gd+ lesions was reduced during the year following the switch to fingolimod (Khatri et al., 2011).

Throughout months 13-24, the beneficial clinical and MRI measures of disease were maintained or improved in patients on continuous fingolimod therapy (Khatri et al., 2011). During the 2-year study course, ARR was significantly reduced by 46% in 0.5 mg fingolimod group compared with either IFNβ-1a to fingolimod switch group (Khatri et al., 2011). More patients in the continuous 0.5 mg fingolimod group were free from new T2 lesions and had significantly fewer new or enlarging T2 lesions compared to the switch group (Khatri et al., 2011). In comparison to the switch group, continuous fingolimod treatment resulted in significantly fewer Gd+ lesions over 24 months and a greater proportion of patients free from Gd+ lesions (Khatri et al., 2011). No significant difference between either continuous fingolimod groups or the switch group was observed in the EDSS score (Khatri et al., 2011).

Considered more efficacious than injectable DMTs, natalizumab and fingolimod are commonly used as second-line therapies for the treatment of RRMS in the event of
treatment failure or intolerability (Kalincik et al., 2015). A prospective cohort study performed by Kalincik et al. (2015) compared the efficacy of switching to natalizumab or fingolimod as a result of injectable DMT treatment failure. Both treatment groups experienced decrease in ARR (Kalincik et al., 2015). The relapse rate was 50% lower for patients who switched to natalizumab than fingolimod (Kalincik et al., 2015). Congruently, a greater proportion of patients on natalizumab were relapse free at the study’s end (Kalincik et al., 2015). No significant difference between the treatment groups was observed in the 6-month sustained disability rates (Kalincik et al., 2015). However the likelihood of experiencing a 6-month sustained regression of disability was 2.8 times greater for patients treated with natalizumab (Kalincik et al., 2015). Based on this study, switching from injectable DMTs to natalizumab was more effective in reducing the rate of relapse and short-term disability in active RRMS (Kalincik et al., 2015).

**Dimethyl fumarate**

Two hundred forty milligrams of dimethyl fumarate (DMF) is administered orally twice daily (Mallucci et al., 2015). Fumaric acid esters modulate the immune response through the production of anti-inflammatory Th2 cytokines (Mrowietz et al., 1998). Beyond its immunomodulatory properties, DMF is believed to have neuroprotective properties.

Based on a study performed by Scannevin et al. (2012), DMF and monomethyl fumarate (MMF), a DMF metabolite, induced the nuclear factor erythroid-derived 2-like
2 (Nrf2) antioxidant response pathway in CNS cells. This study suggested that astrocytes, oligodendrocyte progenitor cells (OPCs), and possibly neurons respond to DMF and MMF, inducing the cytoprotective response (Scannevin et al., 2012). MMF increased intracellular levels of glutathione (GSH), an important antioxidant that prevents damage to cellular components caused by reactive oxygen species (ROS) (Scannevin et al., 2012).

According to the DEFINE and CONFIRM studies, delayed-release DMF significantly reduced new or enlarging T2 hyperintense and Gd+ lesions compared with placebo at the two-year study end (Douglas et al., 2014; Fox et al., 2012). Reductions in the number and volume of both lesion types, as well as the number of T1 hypointense lesions were observed at six months and sustained throughout the trial period (Douglas et al., 2014). Reduction in the volume of T1 hypointense lesions was first observed at one year from the onset of treatment (Douglas et al., 2014).

A significant reduction in the number of patients who experienced at least one relapse in the two-year study period was observed for DMF b.i.d. administration (Gold et al., 2012). Treatment with DMF b.i.d. resulted in a 49% reduction in the risk of relapse (Gold et al., 2012). The ARR at two years was reduced 53% patients treated with DMF b.i.d. (Gold et al., 2012). Compared to 27% of patients taking the placebo, 16% of patients treated with DMF b.i.d. experienced a progression of disability (Gold et al., 2012).

Adverse events, ranging from mild to moderate severity, occurred across all three groups, but with greater frequency in groups receiving DMF (Gold et al., 2012). Most of the adverse events were mild and reversible and did not result in the discontinuation of
treatment. The most frequently reported adverse events included flushing, relapse, diarrhea, nausea, upper abdominal pain, proteinuria, abdominal pain, pruritus, and vomiting (Gold et al., 2012). Flushing and gastrointestinal events occurred most frequently in the first month, tapering off thereafter (Gold et al., 2012). With the exception of relapse, serious adverse events occurred less than 1% of the time (Gold et al., 2012; Fox et al., 2012). In DMF treatment groups, white-cell counts and lymphocytes decreased by approximately 10% within one year, but then plateaued within normal ranges (Gold et al., 2012). Only 4% of patients had white-cell counts less than $3.0 \times 10^{9}$ per liter and lymphocyte counts less than $0.5 \times 10^{9}$ per liter (Gold et al., 2012).

**Teriflunomide**

Approved by the FDA in 2012 for RRMS, oral teriflunomide, an active metabolite of leflunomide, is administered daily at either a dose of 7 or 14 mg (Mallucci et al., 2015). Although its mechanism is not fully elucidated, teriflunomide inhibits mitochondrial dihydroorotate dehydrogenase, which is critical in the de novo synthesis of pyrimidines (Claussen and Korn, 2012). Pyrimidine synthesis inhibition and pyrimidine pool depletion effectively suppresses lymphocyte function (Claussen and Korn, 2012). Additionally, teriflunomide inhibits the interaction between APCs and T cells (Claussen and Korn, 2012). Teriflunomide targets rapidly proliferating immune cells, while preserving the function of slowly dividing cells that utilize exogenous pyrimidine supplies (Claussen and Korn, 2012).
Based on a 108-week study conducted by O’Connor et al. (2011), oral teriflunomide significantly reduced the risk of relapse in comparison with placebo by 31.2% and 31.5% with 7 mg and 14 mg, respectively. Additionally, the TOWER trial confirmed a significant beneficial effect of 7 and 14 mg teriflunomide on the ARR (Confavreux et al., 2014). Both dosages extended the time to the first relapse and increased the number of relapse free patients (O’Connor et al., 2011). The 12 week sustained disability progression did not differ significantly between the placebo group and the teriflunomide groups (O’Connor et al., 2011). Several MRI measures of disease activity were significantly improved by teriflunomide (O’Connor et al., 2011). Significant reductions in T2 hyperintense lesion volume from the baseline were observed for 14 mg teriflunomide (O’Connor et al., 2011). Patients in both active treatment groups had significantly fewer Gd+ lesions (O’Connor et al., 2011). Alterations in brain atrophy were not significantly different among teriflunomide groups and placebo group (O’Connor et al., 2011).

Similar proportions of adverse events occurred among the three study groups (O’Connor et al., 2011). Occurring in greater than or equal to 10% of teriflunomide patients, nasopharyngitis, headache, diarrhea, fatigue, elevated alanine aminotransferase levels, hair thinning, influenza, and pain in arms, legs or back were the most common adverse events (O’Connor et al., 2011). Additionally, pregnancy was treated as an adverse event, since the therapy has not been approved for this population (O’Connor et al., 2011). Two cases of PML were observed during long-term exposure of leflunomide for the treatment of rheumatoid arthritis (O’Connor et al., 2011).
In a head to head study, no significant difference in ARR existed between 14 mg teriflunomide and IFNβ-1a; however, ARR was significantly higher in patients treated with 7 mg teriflunomide (Vermersch et al., 2013). Similar effects on time to failure were observed among the three study groups (Vermersch et al., 2013). It can be concluded that teriflunomide at the higher dose of 14mg/day has an efficacy similar to that of self-injectable DMTs and is appropriate for utilization as a first line therapy. Although FDA approved, the 7 mg teriflunomide is rarely utilized in clinical practice.
III. Regenerative Therapeutics

Immune modulation has been the primary focus of MS medications for the past 20 years. These medications reliably stave off relapse and long-term disability that occurs as a result of relapses but have not been proven beneficial for patients with progressive MS. The clinical benefit of these drugs is undeniable; however this benefit is primarily limited to RRMS. Emerging therapies aim to reverse neurodegenerative damage via OPC stimulation. Remyelinating therapies appear promising for the treatment of all stages of MS.

Stimulation of OPC migration and differentiation is directed by regulatory signals (Bradl and Lassmann, 2009). Certain growth factors, chemotropic molecules, and chemokines, as well as contact-mediated mechanisms, play a role in OPC migration (Bradl and Lassmann, 2009). Differentiation is spatially and temporally regulated through signaling processes such as Notch1, leucine-rich repeat and immunoglobin domain-containing Nogo receptor-interacting protein (LINGO-1), and γ-secretase and other unidentified molecular mechanisms (Bradl and Lassmann, 2009). Highly coordinated, ensheathment of axons occurs during a 12 to 18 hour window early on in OPC differentiation (Bradl and Lassmann, 2009). Oligodendrocytes produce and support membranes, comprise mainly of myelin basic protein (MBP) and proteolipid protein (PLP), as large as 100 times its cell body weight (Bradl and Lassmann, 2009).

In order to produce these necessary proteins, myelinating oligodendrocytes have tremendous metabolic rates and therefore consume large quantities of oxygen and ATP
Hydrogen peroxide and reactive oxygen species are toxic byproducts of this rapid metabolism (Bradl and Lassmann, 2009). Furthermore, myelination requires iron, which under unfavorable conditions promotes the production of peroxides and radical species (Bradl and Lassmann, 2009). A decrease in the fidelity of protein synthesis, a rapid metabolic rate, and high intracellular iron concentrations render oligodendrocytes particularly vulnerable to oxidative damage and mitochondrial injury (Bradl and Lassmann, 2009).

Additional, inflammatory cytokines may cause oligodendrocyte damage or death (Bradl and Lassmann, 2009). TNF-α can stimulate oligodendrocyte apoptosis, while IFNγ is highly toxic for proliferating OPCs and slightly toxic for immature oligodendrocytes (Bradl and Lassmann, 2009).

These mechanisms are promising targets for pharmaceutical development. Numerous compounds are in various stages of clinical development and may prove efficacious for all MS phenotypes. Additionally various approved MS therapies are under investigation for possible regenerative and neuroprotective properties. Below, four therapeutics with reparative potential which are currently being studied, are discussed.

**Monoclonal antibody BIIB033**

LINGO-1, a transmembrane protein expressed in neurons and OPCs, negatively regulates neurite outgrowth and oligodendrocyte differentiation and myelination via the RhoA pathway (Mi et al., 2005). RhoA-GTP mediates actin polymerization and cytoskeleton configuration, and LINGO-1 decreases intracellular levels (Mi et al., 2005).
Inhibition of LINGO-1 promoted myelination and greatly reduced disease severity at all stages in EAE animal models (Tran et al., 2014). Monoclonal antibody BIIB033 binds LINGO-1 with high affinity and specificity (Tran et al., 2014). BIIB033 has a limited ability to cross the blood-brain barrier, but the systemic delivery of high doses has proved efficacious (Tran et al., 2014). In phase I clinical trials, doses up to 100 mg/kg were well tolerated, with headache, upper respiratory and urinary tract infection, nasopharyngitis, and gastroenteritis among the most common adverse events (Tran et al., 2014). No serious adverse events or deaths occurred (Tran et al., 2014). Additionally, only two participants in BIIB033 groups tested positive anti-BIIB033 antibodies, suggesting a low immunogenicity (Tran et al., 2014). BIIB033 is currently in phase 2 trials (Kremer et al., 2015).

*rHIgM22*

Fibroblast growth factor (FGF-2) produced by neuronal cells, astrocytes, and microglia, upregulates platelet-derived growth factor (PDGF) alpha receptor expression on early OPCs (Watzlawik et al., 2013). In conjunction with their receptors, neuronal cells and astrocytes produce PDGF, a potent mitogen, which stimulates early OPC proliferation and promotes survival in OPCs (Watzlawik et al., 2013). More mature cells in the oligodendrocyte lineage are not mediated by PDGF (Watzlawik et al., 2013). This PDGF pathway is considered a possible target for the human monoclonal IgM antibody, rHIgM22.
According to several animal models, rHIgM22 accumulates in lesions and indirectly promotes OPC proliferation and myelination in the presence of PDGF and inhibits apoptotic signaling in OPCs (Warrington et al., 2007; Watzlawik et al., 2013). The monoclonal antibody may increase the sensitivity of OPCs to endogenous repair factors (Watzlawik et al., 2013). These favorable preclinical studies have prompted an investigative phase I clinical trial for use of intravenous rHIgM22 in MS patients (Kremer et al., 2015).

**GNbAC1**

Human endogenous retrovirus type W (HERV-W) occupies up to 8% of the human genome (Kremer et al., 2013). The reactivation of HERV-W may secrete extracellular viral particles that exert a proinflammatory event (Kremer et al., 2013). These particles are strongly associated with clinical disease progression and prognosis (Kremer et al., 2013). According to a study performed by Kremer et al. (2013), the envelope protein (ENV) of HERV-W inhibited the endogenous repair capacity of the CNS. ENV-mediated activation of OPC expressed toll-like receptor 4 (TLR4) and stimulated the production of proinflammatory cytokines, iNOS, and nitrotyrosine groups, which consequently inhibited OPC differentiation (Kremer et al., 2013).

GNbAC1 is a humanized monoclonal anti-ENV antibody (Kremer et al., 2014). It reduces the ENV-mediated proinflammatory cytokine transcription, nitrotyrosine formation, and iNOS induction, making it a promising candidate for a two-pronged therapeutic approach (Kremer et al., 2014). In phase IIa randomized placebo-controlled
clinical study, eight patients received five infusions, one every four weeks, of either 2 or 6 mg/kg intravenous dose of GNbAC1 (Derfuss et al., 2014). GNbAC1 was well tolerated, and the incidence of adverse events did not differ between the two study groups (Derfuss et al., 2014). One serious adverse event was reported, but it was considered to be unrelated to the treatment (Derfuss et al., 2014). Throughout the entire study period, no patient developed antibodies against GNbAC1, suggesting low immunogenicity (Derfuss et al., 2014). With one exception, patients had stable MRI measures of disease progression at six months (Derfuss et al., 2014). This study observed no changes in cytokine profiles, but it can be assumed that changes would only be observed with a longer treatment period (Derfuss et al., 2014). Further investigations into the safety and efficacy of GNbAC1 are schedule to start in 2015 (Kremer et al., 2015).

**Benztropine**

Benztropine is an orally available FDA approved drug that readily crosses the blood-brain barrier and is employed for the management of Parkinson’s disease and dystonia (Deshmukh et al., 2013). It is a centrally acting anti-histamine, a dopamine re-uptake inhibitor, and a cholinergic receptor inhibitor; the latter is believed to produce the primary pharmacological effects (Deshmukh et al., 2013). A study performed by Deshmukh et al. (2013) explored the potential therapeutic effects of benztropine for the treatment of MS.

Benztropine induced differentiation of immature OPCs and remyelination in EAE rodent models (Deshmukh et al., 2013). Benztropine-induced differentiation was
inhibited in the presence of carbachol, suggesting that its mechanism of action is dependent on M1/M3 muscarinic receptor antagonism (Deshmukh et al., 2013). Furthermore, other muscarinic acetylcholine receptor antagonists induced OPC differentiation, but benztropine was the most potent of the tested compounds (Deshmukh et al., 2013). In this study, daily intraperitoneal injection led to functional recovery, significantly decreased clinical severity comparable to or better than fingolimod or IFNβ, and essentially eliminated the risk of relapse (Deshmukh et al., 2013). Benztropine did not influence leukocyte infiltration, prevalence, or activity (Deshmukh et al., 2013). Additionally, the combination of suboptimal doses of benztropine and fingolimod was assessed, with favorable outcomes. Clinical severity was significantly decreased, and benztropine had no adverse effect on the immunomodulatory capacity of fingolimod and visa versa (Deshmukh et al., 2013).

Further preclinical and clinical evaluation of benztropine will be required prior to its use in MS patients. This avenue should be further explored.
IV. Treatment Plans

In the early to mid 90s, the FDA approved the first MS disease modifying therapy, interferon beta. Since then several immunomodulatory and immunosuppressant drugs have been approved. No cure exists for MS. These drugs can partially ameliorate and delay clinical and MRI measures of disease progression, but they are associated with adverse effects and risks. Along with these contraindications, the heterogeneous nature of the disease may obfuscate one’s ability to select the most suitable therapy.

Although subcutaneous and intramuscular injectable DMTs have been widely prescribed for the past two decades, the introduction of intravenous and oral therapies have provided patients with a variety of treatment options. IFNβ and GA have well documented safety profiles with minimal side effects and are increasingly utilized as first-line therapies. RRMS patients generally respond well, and with minimal adverse effects, to the first-generation self-injectable DMTs. Nevertheless, injectable therapies have notoriously low adherence and persistence rates in comparison to other available MS treatments (Wingerchuk and Carter, 2014). While it seems prudent to utilize a time-proven therapy, medications are only effective if taken properly. Furthermore, clinical efficacy and the consequences of chronic inflammation on the CNS should be taken into consideration when devising a treatment plan.

Oral medications provide a convenience that the other MS medications lack. They are easily administered and do not require special handling or training. Three oral
medications, ranging in safety and efficacy, have been approved for the treatment of RRMS: teriflunomide, DMF, and fingolimod.

Once-daily oral teriflunomide, although an effective therapy, does not demonstrate a significant difference compared to IFNβ-1a therapy. Special monitoring is required for its administration due to the risk of hepatotoxicity and a long half-life. Additionally, teriflunomide has not been approved for the use in pregnancy. In the event of overdose, treatment discontinuation, pregnancy, or the desire to become pregnant, teriflunomide’s elimination can be increased with the administration of cholestyramine (O’Connor et al., 2011). With a predominately younger female population, adverse events such as hair loss and potentially teratogenic effects are major deterrents. This medication is perhaps more suitable for patients with intolerances to other medications and those beyond child bearing years. Furthermore, while no direct comparisons have been made, it does not appear that teriflunomide has superior efficacy over the other available MS therapies.

The DEFINE and CONFIRM phase 3 studies illustrated the beneficial effects of twice-daily oral DMF as an immunomodulatory therapy for RRMS. The CONFIRM trial, though not a direct comparison, evaluated the benefit of DMF versus GA and estimated that DMF had similar or greater efficacy to GA across treatment end points (Fox et al., 2012). Treatment with DMF resulted in a greater reduction in ARR (Fox et al., 2012). As well as being an immunomodulatory, DMF is believed to have neuroprotective properties.
Once-daily oral fingolimod has been approved as a first-line therapy for RRMS, but it is often reserved as a second-line therapy. It arguably requires the most intensive laboratory and physical assessments upon treatment initiation. The FREEDOMS and TRANSFORMS trials illustrated the superior efficacy fingolimod compared with IFNβ-1a. As made evident by the prospective cohort study performed by Kalincik et al. (2014), natalizumab has greater beneficial effects on ARR, sustained disability regression, and MRI measures of disease activity, yet natalizumab’s associated PML risk is a reasonable deterrent.

Although lacking direct comparisons, natalizumab is generally considered the most efficacious RRMS therapy available. It is also the most invasive therapy, since it may only be administered intravenously. Regardless of its superior efficacy, natalizumab is associated with an increased risk of developing PML correlating to a prior or new infection with the JC virus. Viral statuses, of patients that start natalizumab therapy with a negative JC virus status, should be check regularly. Furthermore, the wholesale cost of the medication is very expensive, and there are additional costs for infusions and monitoring. This medication is perhaps best suited as a second-line therapy for patients that have experienced treatment failure and disability progression.

Choosing the appropriate therapy is highly complex, with a variety of contributing factors. Medical history, disease aggressiveness, tolerance, adherence, financial burden, quality of life, and lifestyle should all be taken into consideration before treatment application. Treatment strategies may include the utilization of MS drugs in a sequential manner (Figure 4).
Figure 4. Sequential Therapeutic Strategy for MS.

*Fingolimod is approved as a first line therapy but is generally reserved as a second line therapy.
Given that MS is the most prevalent disabling neurological condition among young adults, hundreds of millions of dollars are dedicated for the research and development of new and improved pharmaceuticals. Remarkable progress and innovations in MS therapy have been seen in the past two decades. Preventing disease progression and improving from acquired deficits should be the objective of future therapeutics. With little available for patients suffering from progressive forms of MS, this future research should be directed toward filling this treatment gap.

Emerging remyelinating therapies have yielded promising results in animal models for the treatment of all stages of MS. These drugs could likely be utilized in conjunction with immunosuppressive therapies. Humanized anti-monoclonal antibodies have elicited high levels of research interest. Additionally, antihistamine and anticholinergic compounds have been identified as promoters of OPC differentiation (Kremer et al., 2015). Aside from novel remyelinating therapies, available pharmaceuticals are being reassessed for additional regenerative properties and repurposed.

Personalized DMT strategies are on the pharmaceutical horizon for the treatment of multiple sclerosis. Scientists are making breakthroughs in identifying risk factors, and with the advancements in genomics, answers regarding the etiology of MS may soon be tangible.
REFERENCES


Fox, R. J., Miller, D. H., Phillips, J. T., Hutchinson, M., Havrdova, E., Kita, M., …


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CURRICULUM VITAE

KATHLEEN KAY
Address: 46 W Eagle St. #1, Boston, Ma 02128 • Phone: (217)821-8388
Email: kkay2436@bu.edu • Year of Birth: 1992

EDUCATION
Boston University School of Medicine - Boston, MA
Master of Science in Medical Sciences, 2015 (anticipated)
DePauw University - Greencastle, IN
Bachelor of Arts in Biochemistry, May 2014
  Minor: Biology

PROFESSIONAL EXPERIENCE
The Thomas Shop
Retail Sales Consultant, Summer 2008 and 2010
  • Aided customers with shoe and clothing fittings. Organized window displays and
    floor layout.
Child Care
Child Care Provider, Summer 2010 and 2011
  • Cared for four boys five days a week. Prepared well-balanced meals and
    organized daily activities.

RESEARCH EXPERIENCE
Spring 2013 – Spring 2014
  • Work with Professor Jeffrey Hansen on an aldol epoxidation project. Synthesized
    epoxides and tested their lethality for approximately 8-10 hours a week.

VOLUNTEER WORK
DePauw Student Friend
Student Mentor, August 2013 – May 2014
  • Mentored a four-grade girl with academic and social difficulties. Interacted with
    her weekly at her elementary school.
Organic Chemistry Tutor
Tutor, January 2012 – May 2014
  • Gave private organic chemistry instruction to DePauw University students.
    Taught weekly, unless the student requested extra instruction.
Alpha Chi Omega Foundation
Volunteer, January 2011 – May 2014
• Raised funds for victims of domestic violence. Assisted with awareness seminars and fundraisers. Volunteer at the Women’s Crisis Center in Crawfordsville, IN.

SKILLS
• Laboratory - rotary evaporation, liquid/liquid separation, thin layer chromatography, BSLA, gel electrophoresis, protein crystallization, PCR, cell staining and counting, cell culture
• Computers – Zotero, EndNote

ACTIVITIES/HONORS
2010-2013 • DePauw Women’s Tennis Team
Fall 2010, Spring and Fall 2011 • Dean’s List at DePauw University
2014 • Phi Lambda Upsilon – National Chemistry Honor Society