

REVIEW

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Emerging Therapies for the Management of Multiple Sclerosis

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Abstract

Objective: To provide a comprehensive overview on the emerging treatments used for the treatment management of multiple sclerosis (MS).

Data Sources: PubMed, MEDLINE, Cochrane, and Toxnet databases were used to conduct all comprehensive literature searches over the time period of 1989 to 2009. Search terms such as: multiple sclerosis and oral treatment, monoclonal antibodies, hormonal therapy, and stem cell transplant were used as key word search indicators.

Study Selection: A total of 48 studies were reviewed and selected based on Level 1, 2, and 3 search strategies.

Data Extraction: Level 1 search strategies were initially aimed at evidence-based trials of large sample size ($N > 100$) with a randomized, double-blind, placebo-controlled design in the area of specialized interest. A level 2 search was conducted for additional trials that had many but not all of the desirable traits of evidence-based trials. In addition, a level 3 search strategy was conducted to compare key findings stated in anecdotal reports of very small ($N < 15$), poorly designed trials with the results of well-designed, evidence-based trials identified in level 1 and/or level 2 searches.

Data Synthesis and Conclusion: Despite the wide array of recent treatment advances in the field of MS, the cure still remains elusive. At present, current available treatments at best are only able to slow disease progression by reducing the incidence severity and duration of MS attacks. Recent treatment advances involving the use of newly designed orally administered drugs, monoclonal antibodies with the introduction of stem cell transplantation have revolutionized clinical outcomes for MS patients. Despite great strides made toward disease attenuation, the risks associated with the new treatments are real and have to be weighed against the projected benefits of drug treatment for a disease which has no cure.

Keywords: disease modifying therapy, multiple sclerosis, oral agent, hormonal therapy, monoclonal antibodies, stem cell transplant

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Introduction

Multiple Sclerosis is an inflammatory autoimmune disease of the central nervous system (CNS). The proposed pathophysiological mechanisms of MS are thought to begin when unknown, foreign antigens are introduced into the peripheral blood to induce an immune response. Antigen presenting cells capture and display the foreign antigens to inflammatory T-helper cells (Th1 cells) which subsequently results in their activation. Once activated in the blood, the Th1 cells produce inflammatory cytokines (such as TNF- α , IL-12, and INF- γ) which through a positive feedback cycle further enhance the immune cascade, resulting in the exponential production of inflammatory Th1 cells. The resultant elevated production of inflammatory cytokines from proliferating Th1 cells subsequently results in the up-regulation of adhesion molecules (ICAM-1 and VCAM-1) that line the surface of the blood brain barrier (BBB). Following the upregulation of the adhesion molecules, the circulating Th1 cells can now dock, adhere and aggregate at the surface of the endothelial lining of the BBB, where their continued liberation of inflammatory cytokines facilitate the induction of matrix metalloproteinases (MMP-3 and MMP-9). These metalloproteinases subsequently increase the permeability of the endothelial cells that comprise the BBB. The resultant compromised integrity of the BBB subsequently allows activated Th1 cells to transverse the BBB where they gain direct access into the CNS.¹⁻³ Following migration into the CNS, Th1 cells are then reactivated to specific myelin antigens that share a similar amino-acid sequence homology to that of the original causative antigen that initiated the immune response in the blood. The reactivation of Th1 cells in the CNS results in a similar immune response that originally occurred in the periphery that subsequently drives the phagocytotic activity of additional inflammatory mediators such as B cells, macrophages, lymphokines, and other antibodies that orchestrate a targeted destruction of CNS myelin.^{1,4-6}

There are three main types of MS: relapsing-remitting MS (RRMS), primary progressive MS (PPMS), and secondary progressive MS (SPMS).⁷ Approximately 85% of all MS patients are diagnosed with RRMS.⁸ These patients experience a typical cyclical pattern of clinical attacks followed by periods of complete remission back to normal baseline functioning.

According to the *New Diagnostic Criteria for MS: Guidelines for Research Protocols*, a clinical attack or relapse is any major neurological dysfunction (such as a temporary loss of vision, tingling or numbness in a limb, or muscle spasticity) that lasts for more than 24 hours, often lasting a few days or even several weeks in the absence of elevated body temperature.⁹ In order to diagnose a patient with RRMS, the patient must have experienced at least two clinical attacks or relapses with or without evidence of lesions appearing on the magnetic resonance imaging (MRI) scan. As further specified by the *New Diagnostic Criteria*, “The two attacks must involve different parts of the CNS, must be separated by a period of at least one month, with each attack lasting a minimum of 24 hours”.⁹ Based on this diagnostic criteria, a patient can also be diagnosed with RRMS if the patient has one clinical attack and a MRI exam that fulfills criteria for dissemination in time (additional lesions after 1 month) or space (an increase in the number of lesions).¹⁰ Lesions are also caused by the demyelination of the nerve axons, which can be viewed as white spots on a brain and/or spinal T2 weighted MRI.⁹ Approximately 75% of patients diagnosed with RRMS will progress to SPMS after approximately 12 years.^{11,12} The disease course of SPMS causes a continual and progressive decline in neurological functioning. Patients never completely recover from clinical attacks, and are left with permanent residual neurological deficits with progressive gradual debilitation.^{2,4,7,13} PPMS can be the most debilitating form of MS, however clinical presentation amongst individual patients vary such that some PPMS patients have minimal disability due to the extreme slow nature by which the disease progresses. Fortunately, of the 3 main types of MS it represents only about 15% of MS patients.⁸ PPMS patients do not ever experience a clinical attack but display a continual worsening and progression of symptoms without every recovering or entering into a state of remission. As the PPMS disease course begins right at the onset of the disease, patients are continually accumulating disability and neurological deficit.^{2,7,13-15}

Methods

A review of the literature was conducted utilizing the databases PubMed, MEDLINE, Cochrane, and Toxnet for studies published between 1989 and 2009. The search



term, “multiple sclerosis” was used along with each of the agents mentioned in this review including: cladribine, alemtuzumab, estriol, stem cell transplant, natalizumab, interferon beta-1a, and interferon beta-1b. Key search words such as: oral agent, monoclonal antibodies, hormonal treatments and other variations of these words were used in the search engine to help gather a more comprehensive search result. Evidence based classification of the literature in accordance with Level 1, 2, and 3 search strategies were conducted by the authors of this manuscript.^{9,16} Level 1 search results consisted of evidence based trials of large sample size ($N > 100$) with a randomized, double-blind, placebo-controlled design conducted in the specialty area of interest. To further enhance the quality of information provided in level 1 search results, the PubMed database was searched for meta-analyses pertaining to the various specific areas identified in this paper. Level 2 search results had many but not all of the desirable traits of evidence-based trials, while level 3 results were compiled from anecdotal reports of very small ($n < 15$), poorly designed trials. In some instances, these small trials raised some interesting perspectives that were not addressed in the larger evidence-based trials. Detailed information regarding specific study parameters (P and N values, statistics, patient groupings, assessment criteria, and others) were provided for all pertinent studies. Information on each agent obtained from this search strategy will be discussed in detail and summarized in this review.

Results

Many new emerging treatment strategies have been evaluated for their effectiveness in the management of MS. Forty-eight studies were reviewed and selected for this review. Specifically, the development of drugs such as cladribine, alemtuzumab, estriol and stem cell transplants have all been cited in the scientific literature for managing MS. In addition, new information on existing agents used in the treatment of MS is also discussed. The current review will specifically evaluate their clinical effectiveness and address their recommended placement in MS therapy.

Current Treatment for MS

Unfortunately, there is currently no cure for MS. There are, however, a variety of treatment drugs that have been shown to decrease the frequency, intensity

and duration of relapses in RRMS patients. These treatments may also have the potential to reduce the inflammation and lesions in the brain and improve the quality of life for the patient. Immunomodulatory drugs that act to modify the course of MS include beta interferon 1b (Betaseron[®]), beta interferon 1a (Avonex[®], Rebif[®]), and glatiramer acetate (Copaxone[®]).⁴ The exact disease-modifying mechanism of these drugs is not completely understood. Among other actions, however, these drugs in general decrease the permeability of the BBB and suppress T-cell production, thus preventing the inflammatory cascade from escalating in severity. Natalizumab (Tysabri[®]) has recently been re-entered into the market as second line monotherapy for the treatment of RRMS in patients who have had an inadequate response to, or are unable to tolerate, other therapies. Natalizumab works by blocking the migration of leukocytes into the CNS by preventing $\alpha 4$ -integrin from binding to VCAM-1 on endothelial cells. As a result, this agent has the potential to prevent inflammation from occurring in the CNS by blocking the central migration of blood borne activated Th1 cells. However, there were recent concerns regarding the development of progressive multifocal leukoencephalopathy (PML), a serious opportunistic infection, resulting in this agent's withdrawal in February 2005. Natalizumab was reintroduced into the market in 2007 under strict conditions that have reserved its use as a second line agent in accordance to the above mentioned criteria governing its use.¹⁷⁻²⁰ Other immunosuppressive agents such as methotrexate, cyclophosphamide, and mitoxantrone decrease the activity of the immune system. While they are not frontline therapies, these agents have been recommended for use in specific sub-populations of MS patients.²¹ Mitoxantrone has been recommended for use in patients with RRMS with frequent and incomplete remissions and those patients with SPMS with a rapid progression. Cyclophosphamide should be considered in therapies for patients with a rapid progression of MS who do not respond to less toxic alternatives. Methotrexate may serve as an alternative to other agents to which the patient has exhibited an intolerable toxicity towards. Ultimately, the drug treatment options available for patients with MS are aimed at decreasing the inflammatory immune response in attempt to restore the normal immune system homeostatic balance.



Emerging Treatments for MS

Oral agents

Recent research in the development of an oral disease-modifying agent for MS is actively being pursued with the goal of providing patients with a less invasive mode of treatment administration resulting in improved patient adherence. Some oral agents currently being investigated include cladribine, fingolimod, teriflunomide, temsirolimus, fluoxetine, xaliproden, rosiglitazone, minocycline and simvastatin. Although the scope of this review prevents a detailed comprehensive review of all oral agents being trialed in MS, the recent advances in regard to cladribine, have led to its preferential discussion.

Cladribine

Cladribine is a synthetic anti-neoplastic agent with immunosuppressive properties commonly used in the treatment of hematologic malignancies such as hairy-cell leukemia, chronic lymphocytic leukemia, acute myelogenous leukemia, cutaneous T-cell lymphoma, and non-Hodgkin's lymphoma.²² It also has been evaluated for its potential in the treatment of other autoimmune diseases such as psoriatic arthritis, Sjögren's syndrome, rheumatoid arthritis, and inflammatory bowel disease.^{22,23} It is an analog of the purine nucleoside, adenosine, which plays an important part in DNA synthesis.²² As a result, cladribine interferes with the cellular metabolism and DNA repair, which specifically accumulates in lymphocytes causing programmed lymphocyte depletion while leaving other immune cells unaffected by this agent.²² The use of parenteral cladribine in the treatment of RRMS, SPMS and PPMS has been studied in both Phase II and Phase III trials and has been shown to improve some clinical outcomes such as MRI.^{23,24} Although, the results of clinical outcomes appeared to be not significant, researchers have suggested cladribine may play a role in the earlier stages of inflammation in MS.^{22,23,25,26} The development of an oral cladribine tablet in the treatment of MS was initiated in hopes of providing MS patients with a non-invasive treatment alternative to managing the frequency of relapses. Currently, two trials are being conducted to evaluate the efficacy of oral cladribine 10-mg tablets in the treatment of relapsing-remitting MS.²³ CLARITY (CLAdRibine tablets In Treating MS orally) is a 96 week phase III multicenter, randomized, double-blind, placebo-controlled

trial consisting of 1,327 patients with relapsing-remitting MS from 157 clinical sites internationally.^{23,24,27,28} Patients involved in the CLARITY trial were 18–65 years of age and had received a definite diagnosis of MS according to the McDonald criteria. Furthermore, patients enrolled in the CLARITY trial had RRMS with 1 or more relapses within the past 12 months and no relapse within the past 28 days. Also, a MRI consistent with diagnosis of MS and an Expanded Disability Status Scale (EDSS) of less than 6 were required to participate in the trial. A treatment course was defined as the administration of cladribine tablets once daily for 4 to 5 consecutive days during a 28-day period. In this trial, patients assigned to the treatment group received one of two treatment regimens: 1) two courses for the first and second year; and 2) four courses for the first year and two courses for the second year. The primary efficacy endpoint was defined as the relapse rate at 96 weeks.²³ At the American Academy of Neurology (AAN) 61st Annual Meeting, during a research panel discussion, they provided some interesting results of this agent. A significant reduction in relapse rate was reported and this reduction was shown to be similar for both treatment arms (54.5% for the high dose; 57.6% for the low dose; no P-values reported), compared with placebo. Both dosing regimens also led to a more than 30% reduction in the risk of disability progression (low-dose regimen: $p = 0.018$; high-dose regimen: $p = 0.026$). Progression of disability was measured by a 1-point or greater increase in the EDSS. The number of T1 gadolinium-enhancing lesions from the MRI results was also reduced (87.9% for the high dose; 85.7% for the low dose). However, despite these reported benefits, the risk of neoplasm development was noted in the cladribine arm of the study.

The ONWARD (oral cladribine added ON to rebif new formulation in patients With Active Relapsing Disease) trial was initiated in January 2007 and it is a two-year Phase II multicenter, randomized, double-blind, placebo-controlled study of the safety and efficacy of cladribine tablets.²³ This trial utilized the same dosing schedule as in CLARITY in addition to the administration of subcutaneous interferon beta-1a in patients with RRMS.²³ The primary endpoint of this trial was defined as the safety of this combination therapy at two years. The final results of this study are still pending.



The application of cladribine tablets in MS therapy is advantageous in that only a short course of therapy is required throughout the year. However, patients receiving cladribine must have their blood counts measured prior to receiving treatment, and such monitoring must continue throughout the course of treatment.²³ The adverse effects of cladribine appear to be dose-dependent creating an uncertain toxicity profile with cumulative dosing. In addition, rare idiosyncratic bone marrow suppression resulting in thrombocytopenia, leucopenia, and anemia having been reported at higher doses.²⁹ Moreover, common infections appeared to be a more common adverse effect.²⁷ Furthermore, since MS is disease that predominantly affects females, clinicians should be aware of its potential to cause breast cancer.³⁰ This medication appears to be well tolerated at lower doses.²³

Alternative oral agents in the treatment of MS include fingolimod, laquinimod, teriflunomide, and fumaric acid. Fingolimod is a structural analogue of sphingosine which interferes with cellular traffic between lymphoid organs and blood. It acts as a high affinity agonist responsible for the down regulation of its receptor on lymphocytes, which ultimately leads to a reduction in the recirculation of autoreactive T-cells between the lymph nodes and the CNS.³¹ In a randomized, double-blind, placebo-controlled phase II study, 281 patients with RRMS received either placebo, fingolimod 1.25 mg/day, or fingolimod 5.0 mg/day for 6 months. This was followed by an 18 month extension phase in which all patients were re-randomized to receive either one of the fingolimod treatments. While the results obtained in this study warrant further evaluation of the use of fingolimod in MS, the major limitation of fingolimod is concerned with the frequency and severity of side effects observed in the cohort receiving fingolimod 5 mg/day.³¹ Laquinimod is a new quinolone carboxamide which has demonstrated efficacy in animal models of several autoimmune diseases, including MS.³² While it does not lead to immunosuppression, it likely exerts its immunomodulatory effects through a Th1/Th2 shift. In a phase II, randomized, double-blind, placebo-controlled 36 week study, the efficacy and safety of laquinimod was evaluated.³¹ 102 subjects received placebo, 98 subjects received laquinimod 0.3 mg twice per day, and 106 subjects received laquinimod 0.6 mg twice per day. The use of 0.6 mg

laquinimod demonstrated a significant treatment effect in the reduction of MRI activity ($p=0.0048$) compared to placebo, as did the use of 0.3 mg laquinimod ($p=0.6740$) compared to placebo. The main limitation in the use of laquinimod is concerned with the safety profile of the drug. Laquinimod may potentially result in hepatotoxicity or a possible pro-inflammatory response. Four major side effects reported in the phase II study include: pleuritis, Budd-Chiari syndrome, hemorrhagic pituitary adenoma, and the possible development of Chron's disease.³¹ Teriflunomide is the active metabolite of leflunomide. The ability to noncompetitively and reversibly inhibit the mitochondrial enzyme dihydro-orotate dehydrogenase, relevant for the *de novo* synthesis of pyrimidine, is believed to exert the most important therapeutic effect.³³ In a recent randomized, double-blind, placebo-controlled phase II study, 179 patients with MS (RRMS $n = 157$, SPMS $n = 22$) received either placebo, teriflunomide 7 mg/day, or teriflunomide 14 mg/day. Treatment with either teriflunomide regimen resulted in reductions of MRI activity compared to placebo.³¹ Furthermore, the higher dose of teriflunomide appears to have increased effectiveness compared to the low dose regimen. The limitation in the use of teriflunomide is concerned with serious adverse effects including: elevated liver enzymes, hepatic dysfunction, neutropenia, trigeminal neuralgia, nausea, increases in alanine aminotransferase levels, diarrhea, and back and limb pain.^{31,33} The exact mechanism of action of fumaric acid is still unclear. However, *in vitro* experiments have shown that it can influence the expression of cytokines and adhesion molecules which are thought to be involved in the inflammatory cascade.³¹ A phase II, randomized, placebo-controlled, double-blind study evaluated the use of fumaric acid in MS. During the one year study, subjects either received placebo or 720 mg fumaric acid per day for the initial 24 weeks of the study. In the final 24 weeks, all subjects received fumaric acid. A reduction in MRI activity was observed when treated with fumaric acid compared to placebo. The limitation of fumaric acid in the treatment of MS involves the lack of knowledge regarding the exact mechanism of action; as well as reported adverse effects including: nasopharyngitis, headache, nausea, diarrhea, fatigue, and upper abdominal pain.³¹



Monoclonal antibodies

The research that has been conducted on monoclonal antibodies has been increasingly important in the treatment of MS. Its ability to target specific antigens to elicit specific outcomes makes these agents an important alternative for patients living with MS. Natalizumab in the treatment of MS has already entered the market and has been previously discussed. Three other agents are currently in clinical trials and they include: alemtuzumab, daclizumab, and rituximab.^{34–36} In this review, alemtuzumab is discussed in detail.

Alemtuzumab

Alemtuzumab is a humanized monoclonal antibody (mAb) that targets the CD52 antigen of T and B lymphocytes, monocytes, and macrophages.^{37,38} This agent was initially developed for the treatment of lymphoid malignancies, but recent trials have demonstrated its potential role in the treatment of MS.³⁹ In a trial of 58 patients with RRMS (n = 22) or SPMS (n = 36), alemtuzumab (named, Campath-1H, in study) was administered to evaluate the efficacy alemtuzumab in MS treatment.³⁴ Patients in the SPMS group were diagnosed on an average of 11.2 years ago (SD ± 6.1 years) at the time of treatment, of which 3.6 years (±2.6 years) had been in the progressive phase. The mean EDSS score of these patients was 5.8 (±0.8, range 3.5 to 7.0). Patients were recruited if they had experienced an increase in disability in the year prior to treatment of at least one EDSS point, during which annual relapse rate was shown to be 0.7/patient/year. These patients were treated from either 1991 to 1993 or from 1994 to 1997.³⁴ Patients in the RRMS group received alemtuzumab if they were not successfully treated with approved agents or if they have experienced a high relapse rate early in the disease, which may indicate a poorer prognosis. Patients in this group were diagnosed from 9 months to 12 years ago (mean 2.7 years ±2.9 years) at the time of treatment. The mean EDSS score was 4.8 (±2.0, range, 1.0 to 7.5) and had an overall annual relapse rate of 2.2 per patient. Patients were monitored every 3 to 6 months for the first three years post treatment, and then yearly thereafter. Alemtuzumab was administered through a four-hour intravenous infusion of 20 mg once daily for five days. Re-treatment was provided after 12 to 18 months of a fixed dose of 60 mg over three consecutive days

(20 mg per day). The administration of intravenous methylprednisolone of 1 gram over 1 hour was given prior to the alemtuzumab dose to reduce the symptoms of cytokine release syndrome. Results showed that alemtuzumab was able to suppress inflammation in the SPMS group, but its ability to protect patients from disease progression was not demonstrated. After one year of treatment, 91.7% (n = 33) patients in the SPMS group maintained their pre-treatment EDSS score. However, at 8.5 years (±0.5), only 11.1% (n = 4) showed no sustained worsening of disease from their pre-treatment EDSS score. As a group, the mean rate of increase in disability after treatment was +0.2 EDSS points/patients/year, with a significantly reduced rate of progression compared to the year before treatment (p < 0.001). Significant decrease in gadolinium-enhancing lesions followed by increased brain and spinal cord atrophy was observed in the 25 patients from the SPMS group that were able to receive MRI measurements. However, significant reduction in disability was seen in RRMS patients.

In a phase II, randomized, blinded study of 334 patients with RRMS for 3 years or less and an EDSS score of 3.0 or less, alemtuzumab was compared to IFN-beta 1a.⁴⁰ Patients received either alemtuzumab 12 mg/day or 24 mg/day, given for 5 consecutive days during the first month and for 3 consecutive days at months 12 and 24, or interferon beta-1a 44 mg three times per week. The primary endpoints of this study included the time to sustained accumulation of disability, which was defined as an increase of at least 1.5 EDSS points for patients with a baseline score of 0 and of at least 1.0 point for patients with a baseline score of 1.0 or more, and the rate of relapse. In this study, alemtuzumab was shown to have a more significant effect on decreasing the rate of sustained accumulation of disability when compared to interferon beta-1a (9.0% vs. 26.2%; p < 0.001). Moreover, the annual relapse rate was also significantly reduced (0.10 vs. 0.36; p < 0.001). The mean disability score also improved on a 10-point scale of 0.39 point in the alemtuzumab group, and worsened by 0.38 point in the interferon beta-1a group (p < 0.001). The lesion burden on T2-weighted MRI was reduced in the alemtuzumab group compared to the interferon beta-1a group (p = 0.005). These results also emphasized the importance of initiating treatment as early as possible where it is most effective during the inflammatory stage of MS.



The most common adverse effects of alemtuzumab include early infusion-related effects such as fever, headache, rash, nausea, vomiting and rigor, which may be due to cytokine release. This may be managed by administering glucocorticoids, diphenhydramine, or acetaminophen prior to infusion with alemtuzumab. The most common adverse effect of monoclonal antibodies that reduce the number of T-cells include an acute cytokine release syndrome, which is characterized by fever, headache, malaise and urticarial rash. However, it is important to note that 23% of patients receiving alemtuzumab in the phase II trial conducted by the CAMMS223 Trial Investigators developed a thyroid disorder (e.g. Graves disease) vs. 3% in the interferon beta-1a group. Immune thrombocytopenic purpura (ITP) was also reported (3% vs. 1% in interferon beta-1a), which resulted in the suspension of alemtuzumab treatment in September 2005 when three patients developed this condition in which one patient died. As a result, frequent blood count measurements must be monitored. Infections also occurred in 66% of patients treated with alemtuzumab (vs. 47% in interferon beta-1a group). Overall, alemtuzumab may be appropriate for use in MS therapy however further studies are currently being conducted which will assist clinicians in determining its place in the MS treatment strategies.

Other monoclonal antibodies in the treatment of MS include daclizumab and rituximab. Daclizumab is a humanized monoclonal antibody which targets the IL-2 receptor on activated T-cells, and blocks the IL-2 alpha chains CD25 of activated T-cells.⁴¹ A phase II, randomized, double-blind, placebo-controlled study known as the CHOICE trial investigated the use of daclizumab in MS patients who were receiving interferon beta therapy. The CHOICE study tested daclizumab in 230 patients with RRMS, divided into three cohorts.⁴¹ Subjects received placebo, daclizumab at a high dose, or daclizumab at a low dose. A significant reduction in lesions, as well as a reduction in the number of relapses was reported in the high dose cohort. These observations indicate that daclizumab may be a promising option for MS patients. While daclizumab is generally well tolerated and no severe side effects have been reported, the lack of knowledge concerning the long-term side effects associated with daclizumab limits its widespread use.⁴¹ Furthermore, the efficacy of daclizumab is likely reduced by the

development of neutralizing antibodies. Rituximab is a monoclonal antibody which targets the CD20 antigen expressed by B-cells.⁴¹ In targeting the CD20, rituximab depletes B-cells from the peripheral blood, and the amount of B-cells, and consequently T-cells, in the cerebrospinal fluid is decreased. In a placebo-controlled, phase II study investigating the use of rituximab in RRMS patients, the cohort receiving rituximab demonstrated a reduction in both lesions and relapses compared to the placebo cohort. Another trial considered the use of rituximab in patients with neuromyelitis, which may represent a subgroup of MS.⁴¹ In this trial, patients with neuromyelitis showed a reduction in the frequency of relapses when treated with rituximab. Despite the loss of B-cells in the peripheral circulation, an increased risk of infections has not been observed. The limitations to the use of rituximab are similar to those of daclizumab. This includes the lack of knowledge regarding the long-term side effects, as well as the production of neutralizing antibodies against rituximab, possibly reducing the effectiveness of the therapy.

Hormonal therapies

Clinical experience and limited research has suggested a potential beneficial neuroprotective and immunomodulatory effect of pregnancy in MS via hormones such as estriol.⁴² However, the reality of the term neuroprotection is often loosely used by clinicians and is a very difficult concept to define in specific regards to a disease such as MS whose mystery involves complex immunopathogenetic mechanisms.⁴³

Estriol

Estriol is an estrogen produced by the fetal placental unit, the presence of which increases steadily during pregnancy. In non-pregnant women, estriol is not present in any substantial amount. The beneficial effect of estriol during pregnancy especially during the third trimester has been observed in MS and other autoimmune diseases including rheumatoid arthritis.⁴² The increase in estriol levels during pregnancy has been demonstrated through *in vivo* studies to ameliorate experimental autoimmune encephalomyelitis (EAE). Additional support for this novel treatment strategy also comes from *in vitro* studies that demonstrated a quiescent effect on activated human T-cells derived from MS patients following the administration of estriol.⁴² As a



result, it has been speculated that during pregnancy, there is a naturally occurring immune shift involving a decrease in inflammatory Th1 cells, and an increase in anti-inflammatory Th2 cells.

Currently, a phase II combination trial is in progress examining the use of estriol in female patients with RRMS. The study is a double blind, placebo controlled, randomized, multicenter trial of oral estriol. This study compares the use of glatiramer acetate combined with either 8 mg of oral estriol per day or placebo over the course of 24 months.^{44,45} The primary outcome is the measurement of relapse rate. The trial involving the recruitment of 150 patients began in 2007 and is anticipated to be concluded in 2012. Other clinical trials are also underway to further determine the efficacy of estrogen treatment in MS.⁴⁵

For example, in a phase I, crossover trial, 12 non-pregnant female patients with clinically defined MS were enrolled.⁴⁶ Six women had RRMS, and six had SPMS. Patients were eligible if they were between the ages of 18 and 50, had an EDSS score of below 6.5, had not received a steroid treatment within the past 3 months, and had not been receiving IFN- or glatiramer acetate for at least 6 months. The trial consisted of a 6 month pre-treatment phase, a 6 month treatment phase with 8 mg estriol per day, and a 6 month post-treatment phase. In the RRMS cohort, there was also a 4 month retreatment phase, which involved the administration of estriol in combination with 100 mg per day of progesterone in order to protect against the possibility of developing endometrial hyperplasia. The objective of this study was to attempt to recreate the beneficial effect of pregnancy in MS patients. All six RRMS patients finished the entire year and a half trial, while only four SPMS patients completed the trial.

Serum estriol levels during both the treatment and retreatment phase were similar to those of pregnant females in their third trimester. Immune responses in delayed type hypersensitivity to tetanus were found to be significantly decreased ($p = 0.006$) at the end of the treatment phase compared to pre-treatment baseline. Levels of IFN- were significantly decreased ($p = 0.003$) in the RRMS cohort after the treatment phase, while there was no decrease in the IFN- of the SPMS patients. The number and volume of T1-weighted MRI enhancing lesions for all 10 patients decreased during the treatment phase, although this due in large

part to the results of the RRMS patients. Following the treatment phase, RRMS patients median total enhancing lesion volumes were decreased by 82% ($p = 0.01$) and the number there of was decreased by 82% ($p = 0.02$). While the volume and number of lesions returned to near pre-treatment baseline levels post-treatment, the 4 month retreatment phase saw a repeated decrease in lesion volume by 88% ($p = 0.008$) and a decrease in number by 48% ($p = 0.04$). While reports of clinical relapses were limited, the EDSS and 9-hole peg test showed no significant changes during the study. Cognitive tests were slightly improved in the RRMS cohort ($p = 0.04$), but not in the SPMS cohort. These results indicate a greater potential benefit from the use of estriol for RRMS patients as compared to SPMS patients.

The use of estriol was well tolerated with the exception of three patients reporting menstrual cycle abnormalities. Endometrial biopsies were performed for these patients, and they came back negative for hyperplasia. In the long term, estriol should be combined with progesterone, similar to the dose administered in the retreatment phase, in order to protect against the risk of developing hyperplasia. Furthermore, the safety profile of estriol compares favorably with currently available immunomodulatory therapies for MS.⁴⁵ Also, it should be noted that in another study, estriol treatment in EAE was found to be non-gender-specific.⁴⁷ The estrogen receptor expressions in both genders were similar, and the severity of EAE was decreased in both females and males when estriol was given compared to placebo.

Stem cell transplants

MS has garnered increased consideration as a potential disease for treatment with stem cell transplantation. Such treatment is based upon an attempt to exploit the effects of immune suppression to regenerate a new and healthy immune system, also referred to as "immune resetting".⁴⁸ Hematopoietic stem cell transplantation (HSCT) can be performed using either autologous or allogenic approaches. This review will focus on autologous HSCT in detail.

Autologous hematopoietic stem cell transplantation

Much of the early focus on HSCT in MS involves autologous methods using stem cells from within that



individual (AHSCT). While using stem cells from a leukocyte antigen matching donor (allogenic) is more attractive compared to autologous HSCT, the associated increased risk of morbidity is far too great. The premise of AHSCT is based upon intense immune suppression in order to eradicate autoimmune cells followed by the re-infusion of previously acquired stem cells which reconstitute the immune system in a self-tolerant manner. AHSCT methods may follow one of two courses of therapy—myeloablative or non-myeloablative.^{49,50} During myeloablative procedures, stem cells such as CD34+ stem cells are mobilized using a monotherapy of granulocyte colony stimulating factor (G-CSF) or a combination therapy of G-CSF and cyclophosphamide, then harvested from the peripheral blood. Stem cells are then conditioned using a regimen with sufficient lympho- and myeloablative properties such as the BEAM method (300 mg/m² BCNU on day 1, 2 × 100 mg/m² etoposide and arabinosylcytosine on days 2 through 5, and melphalan 140 mg/m² on day 6) commonly used in AHSCT in the treatment of cancer.⁴⁹ The stem cells are then re-infused using graft techniques at least 24 hours after the conditioning process. Non-myeloablative techniques involve stem cell mobilization using 2 g/m² intravenous cyclophosphamide, followed by 10g filgrastin daily. The conditioning process consists of 200 mg/kg cyclophosphamide along with 6 mg/kg anti-thymocyte globulin (ATG).⁵⁰ The use of ATG is warranted in order to prevent thrombocytopenia. Recent immune resetting studies have observed the generation of a new and diverse T cell repertoire (TCR), which is significant in the attempt to eradicate the immune system of self-reactive cells. The ability to reset the immune system with an AHSCT facilitates the ability of the newly transplanted stem cells to release of both anti-inflammatory immunomodulatory factors, as well as trophic and growth factors which enable the stem cells to proliferate, differentiate, and migrate to the sites of myelin damage.⁵¹

As a result, various early stage clinical trials have begun to explore the potential merit of AHSCT. For example in a recent phase I/II randomized, mitoxantrone controlled study of non-myeloblastic AHSCT 21 RRMS patients who have had 2 corticosteroid-treated relapses within the previous 12 months, or one relapse and gadolinium-enhancing lesions

seen on MRI separate from the relapse were treated between January, 2003, and February, 2005.⁵² The primary outcomes were progression free survival and reversal of neurological disability at three years post-transplantation. 17 (81%) of patients improved their EDSS score by at minimum one point. None of the patients finished the study with an EDSS score following HSCT lower than that at pre-treatment baseline ($p < 0.0001$). Other significant improvements in neurological functioning were observed, as indicated by improvements in neurological rating scale score ($p = 0.0001$), paced auditory serial addition test ($p = 0.014$), 25-foot walk ($p < 0.0001$), and quality of life as measured with the short form-36 questionnaire ($p < 0.0001$). While 5 patients (24%) relapsed at a mean of 11 months post-treatment following initial improvements, they were treated with further immunosuppressants. After a mean 3 year follow-up, relapse-free survival was 100%.

A larger scale trial such as the Autologous Stem Cell Transplantation International Multiple Sclerosis Trial (ASTIMS) has been established in an attempt to further explore the benefits and risks associated with AHSCT. It is a multicentre, prospective, randomized, phase III trial with clinical patient evaluation at 6 month intervals and MRI patient evaluations at 12 month intervals.⁵³ The mobilization of stem cells involves subcutaneous cyclophosphamide therapy followed by filgrastim therapy five days after the last dose of cyclophosphamide. The conditioning regimen begins 20–40 days after mobilization and consists of the BEAM method combined with ATG. The primary aim is to compare the efficacy and safety of immunoblation and AHSCT with mitoxantrone therapy.⁵³ In a 5 year follow-up of the high-intensity regimen Canadian MS BMT study using either busulfan alone or busulfan combined with cyclophosphamide, none of the 25 treated patients had experienced any further attacks or MRI activity after a mean period of 5.5 years.⁴⁹

Side effects common to AHSCT include mortality; veno-occlusive liver disease; septicemia; therapy induced leukemia, lymphoma, and cancers; the necessity for platelet infusions; premature ovarian and gonadal failure; as well as the long term use of antibiotics, antivirals, antifungals, and immunoglobulin transfusions. Furthermore, additional toxicities also include that of fever and UTIs, as well as accelerated



short-term brain volume loss. However, long term follow-ups have indicated a much lower rate of brain atrophy after the second year of follow-up than previously observed short term.⁴⁸ The greatest concern for AHSCT is transplantation related mortality (TRM). For a disease such as RRMS, TRM should nowadays be below 1%. Reports of TRM from 1995 to 2000 were 7.3%, while it has dropped to 1.3% from 2001 to 2007.⁴⁹ Improvements in patient selection, as well as improvements in clinical techniques are likely involved in the reduction of TRM. Clearly, there is a need to balance efficacy with safety in determining which HSCT therapy regimen should be used for MS patients.

The greatest controversy in HSCT involves the potential use of pluripotent embryonic stem cells. While there are moral and ethical issues to consider, clinical approaches to the use of embryonic stem cells are currently absent. As a result, the focus on stem cell transplantation involves mesenchymal stromal cells (MSC) found in the bone marrow, which are already utilized in other disease states including cancer treatment. MSCs may provide the greatest benefit to younger patients at an earlier stage of MS while the disease is still in the inflammatory stage prior to its progression to the neurodegenerative stage. While further larger studies are needed, there is compelling evidence that AHSCT has the capacity to lead to a complete and sustained cessation of inflammatory activity in patients with MS.⁴⁹

Discussion

Treatment strategies of MS are continuously evolving in efforts to improve patient adherence and overall quality of life for patients living with MS. The treatments currently available function only to slow the disease progression at best and mitigate symptoms. At present, the majority of treatment advances have been made in the RRMS population.

Oral agents such as cladribine have shown some promising results with the potential to provide a less invasive alternative for patients uncomfortable with receiving frequent injections. Cladribine has been shown to exhibit its anti-inflammatory properties by specifically targeting the depletion of lymphocytes. As a result, this agent may hold promise in MS treatment since inflammation is an early component of this autoimmune disease. Although cladribine

appears to produce improvement in relapse rates, overall disability and MRI results in individuals with RRMS, researchers believe cladribine may play a more significant role in attenuating inflammation during the earlier stages of MS. The results from the CLARITY trial support this concept of use in the early stages of MS as the patients enrolled in both treatment arms reported low disease activity. During the early stages of MS, the inflammatory aspects of the disease represent a significant contribution to the display of the early clinical deficits associated with this autoimmune insult. Since the patients enrolled in the two treatment arms with low disease activity, it is indicative that these patients were in the early inflammatory stages of the disease (hence favorable responders) prior to any permanent deficits associated with un-repairable demyelination that are characteristic of later disease stages. Furthermore, due to the mechanism of action of cladribine, risk of opportunistic infection, although rare, is still probable and it is therefore necessary for patients to undergo frequent blood tests during treatment. Patients on oral cladribine are likely to receive either two or four, 5-day courses per year, resulting in a maximum of only 10 or 20 days of therapy per year. Not only does this short and intermittent dosing regimen have the ability to improve patient adherence and quality of life, it may also hold promise in its use as an add-on therapy to available disease-modifying therapies currently in practice.²⁴ However, the use of cladribine as add-on therapy is still being evaluated. Given its current safety and efficacy profile, the use of cladribine may offer future promise to MS patients suffering from this chronic illness.

Monoclonal antibodies, such as alemtuzumab, represent significant advances made in terms of MS treatments due to their ability to target specific antigens thought to be involved in driving the pathological immune response against CNS myelin. Although adverse effects, such as flu-like symptoms are known to be associated with its use, current clinical trials were able to demonstrate the efficacy of alemtuzumab in reducing MRI lesions and improving disability status was shown in both SPMS and RRMS patients. Relapse rate was also shown to be significantly less in RRMS patients. Alemtuzumab may be considered as an alternative therapy for MS in patients who were not successfully treated with current disease-modifying agents or if they have experienced a high



relapse rate early in the disease. Alemtuzumab is administered through a four-hour intravenous infusion of 20 mg once daily for five days. Re-treatment can be provided after 12 to 18 months of a fixed dose of 60 mg over three consecutive days (20 mg per day). The administration of intravenous corticosteroids may be necessary prior to the alemtuzumab dose to reduce the symptoms of cytokine release syndrome. The most common adverse effects of this agent may also include thyroid disorder and ITP. Based on the literature, alemtuzumab may be appropriate for use in MS therapy and further studies are currently being conducted.

The research interest in hormonal agents in MS is continually rising. Hormonal therapies are thought to have both immunomodulatory and neuroprotective properties, which have been shown to have some potential for use in the treatment of MS. This therapeutic concept was developed from the apparent protective effects of pregnancy in MS. Interestingly, estriol treatment in EAE was found to be non-gender-specific. Current trials have evaluated the efficacy and safety profile of oral estriol in the treatment of MS administered estriol at a dose of 8 mg per day. In order to counteract the development of endometrial hyperplasia associated with estrogen treatment, progesterone has been recommended at a dose of 100 mg per day. Estriol was shown to have significant potential in reducing inflammatory mediators as well as MRI lesions in both SPMS and RRMS patients. However, the results of clinical outcomes indicate a greater potential in the use of estriol in the treatment of RRMS than for SPMS. Overall, estriol appears to be well tolerated with the most common adverse effect being menstrual cycle abnormalities. More studies may be warranted before this product can be recommended.

Despite increased patient interest to trial stem cell transplantation, the evidence demonstrating its benefits outweighing the risks is lacking. Although this method of treatment may have the potential to eliminate future relapses and improve the overall quality of life of patients living with MS, mortality, as well as the risk of therapy-induced leukemia, lymphoma, and cancers, make this treatment strategy unfavourable. Due to these limitations, the use of stem cell transplantation is not recommended at this time. However, ongoing studies may offer future advancements in

this technology and provide a better understanding of potential treatment alternatives for MS.

Conclusion

Despite all the research dedicated to this disease, there is still no cure. The treatments currently available function only to slow the disease progression at best and mitigate symptoms. There are a number of therapies that have generated increased interest and potential in the management of MS. Oral agents, monoclonal antibodies, hormonal therapies and stem cell transplantation are treatment strategies that are currently and continuously being evaluated through clinical trials. These emerging treatment strategies may have a role in improving patient adherence, enhancing quality of life, and decreasing the severity and/or frequency of relapses and progression of MS. However, it is still important to consider the risks and potential adverse effects of each agent before recommending its use. Although, advances have been made in terms of designing a suitable and efficacious oral agent, the injectable immunomodulatory drugs such as interferon β 1a (s.c), interferon β 1b, glatiramer acetate and interferon β 1a (i.m) still remain the 1st line treatments for RRMS with natalizumab reserved as second line treatment for those patients that are not responsive or intolerable to the frontline agents. Henceforth, there is a real need for further development of controlled evidence-based trials involving these proposed alternatives surrounding their use in MS.

Abbreviations

BBB, Blood brain barrier; CNS, Central nervous system; EBV, Epstein-Barr virus; ICAM-1, Intercellular adhesion molecule 1; VCAM-1, Vascular cell adhesion molecule 1; IL-2, Interleukin 2; IFN, Interferon; INF- γ , Interferon gamma; MMP, matrix metalloproteinases; MRI, Magnetic resonance imaging; MS, Multiple sclerosis; PNS, Peripheral nervous system; PPMS, Primary progressive multiple sclerosis; RRMS, Relapsing-remitting multiple sclerosis; SPMS, Secondary progressive multiple sclerosis; TCA, tricyclic antidepressants; Th1, T-helper 1; Th2, T-helper 2; TNF- α , Tumor necrosis factor alpha; UTI, Urinary tract infection; PML, progressive multifocal leukoencephalopathy; AAN, American Academy of Neurology; eod, every other day; HSCT, Hematopoietic stem cell transplantation; AHSCT, Autologous hematopoietic



stem cell transplantation; G-CSF, granulocyte colony stimulating factor; BEAM, BCNU-etoposide-arabinosylcytosine-melphalan; ATG, Anti-thymocyte globulin; TCR, T cell repertoire; ASTIMS, Autologous stem cell transplantation international multiple sclerosis trial; MSC, mesenchymal stromal cells.

Disclosures

Christine Leong, Michael Prout, Josee-Anne Le Dorze, Mike Limerick, Emma Frost do not have any conflicts of interest to disclose and have not received any honoraria nor have conducted any clinical trials with any pharmaceutical company that produce immunomodulatory treatments for MS. Dr. Michael Namaka has received honoraria as an invited speaker and participant as a medical advisory board member. Dr. Namaka is involved in various types of MS research and clinical trials that are funded in whole or in part by various pharmaceutical companies such as Boehringer, Pfizer, Serono, Biogen, Bayer, Valeant, Teva, Janssen Ortho. Dr. Farid Esfahani has received honoraria as an invited speaker and participant as a medical advisory board member. Dr. Esfahani is involved in various types of MS research and clinical trials that are funded in whole or in part by various pharmaceutical companies such as Pfizer, Serono, Biogen, Bayer, Valeant, Teva.

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