Chapter 14
Stem Cells for Multiple Sclerosis

Pamela Sarkar and Neil Scolding

Abstract Multiple sclerosis (MS) is a common cause of progressive neurological disability, particularly affecting young adults, and no currently available therapies have any clinically meaningful impact in reversing, halting, or even slowing progression. Stem cell therapies have for several decades held out the prospect of addressing this major therapeutic challenge. Classical *cell replacement* approaches envisaged transplanting stem cells to replace lost oligodendrocytes and to remyelinate denuded axons in focal MS lesions. However, the prominent role of diffuse axonal damage in generating progressive disability limits the applicability of this strategy. A second disparate approach to stem cell therapy in MS is to use autologous *hematopoietic stem cells*, aiming to regenerate the subject's dysfunctional immune system and halt inflammatory damage. Finally, what we have termed *restorative cell therapy* aims to exploit the multiple reparative and/or disease-modifying capacities of autologous mesenchymal or other cell populations, principally from the bone marrow, but potentially from alternative tissues (such as fat), to limit and reverse tissue damage in multiple sclerosis.

Keywords Multiple sclerosis • Remyelination • Cell therapy • Neurodegeneration • Mesenchymal stem cells

Introduction

Multiple sclerosis (MS) affects some 2.5 m people worldwide, principally young adults. It carries an economic burden of around $10 billion annually in the USA and 9 billion euros across the European Union, these costs largely representing the direct and indirect consequences of progressive disability. Most patients start their disease course with a relapsing-remitting presentation, that is, with good recovery after attacks, but over 80 % of patients ultimately develop progressive disability,

P. Sarkar, MBBS, MRCP • N. Scolding, PhD, FRCP (✉)
University of Bristol Learning and Research Building, Southmead Hospital, Southmead Way, Bristol BS10 5NB, UK
e-mail: pamela.sarkar@bristol.ac.uk; N.J.Scolding@bristol.ac.uk

M.H. Tuszynski (ed.), *Translational Neuroscience*,
DOI 10.1007/978-1-4899-7654-3_14

with a median time to progression of some 15 years. Immune-based treatments have proved increasingly effective in reducing relapse frequency, but none so far has been shown to have a meaningful clinical impact in reversing, halting, or even slowing progressive disability. Developing such therapies and targeting established progression is therefore a major healthcare priority. Cell therapy may offer one possible solution.

The question of stem cell therapy for multiple sclerosis has evolved considerably over the past decade and has now acquired considerable complexity. The development of *cell* therapy for MS arguably became realistic almost 40 years ago—long before the explosion of interest in stem cells—with the work of Bill Blakemore in Cambridge [1]. In a remarkable initial experiment, Blakemore showed that exogenous myelinating cells (Schwann cells in this instance), injected into demyelinated lesions in the central nervous system (CNS), achieved successful remyelination. Proof of the therapeutic principle of replacing oligodendrocytes damaged by MS disease processes with healthy (re)myelinating cells was thereby offered. Years later, the major problem of identifying the best candidate remyelinating cell type appeared to have been solved by the emergence of stem cells [2, 3], but, paradoxically, highly informative contemporaneous studies of the clinical biology of MS cast no little doubt on the underlying basis of replacement cell therapy as a treatment approach to this disease [4, 5]. At the same time, other sources and types of stem cell came into focus and alternative ways of exploiting their properties emerged. In consequence, it can now be argued that stem cell therapy in MS is thought of in three quite different ways:

- "Classical" stem cell therapy—aiming to use stem cells in a "cell replacement" strategy, to repair CNS myelin;
- Hematopoietic stem cell therapy—aiming in effect to replace or reset the subject's misfiring immune system, in order to prevent future CNS inflammation; and
- "Restorative" stem cell therapy—utilizing complex additional properties of certain stem cell types with the combined aims of both limiting nervous tissue damage and promoting endogenous tissue regeneration and repair.

These three approaches clearly have different (if overlapping) aims and make use of different types of stem cells. Partly because of this, many now prefer the term "cell therapy" to "stem cell therapy." The three strategies are at different stages of development—some nearer and some further from the clinic. All, as we hope will become clear, are important.

"Classical" Stem Cell Therapy: Replacing Lost Oligodendrocytes for Myelin Repair (Fig. 14.1)

As far as this author is aware, there has only been a single clinical experiment, not formally published [6], exploring this approach in (two) MS patients, and this studied the safety and feasibility of (autologous) Schwann cells, not stem cells.

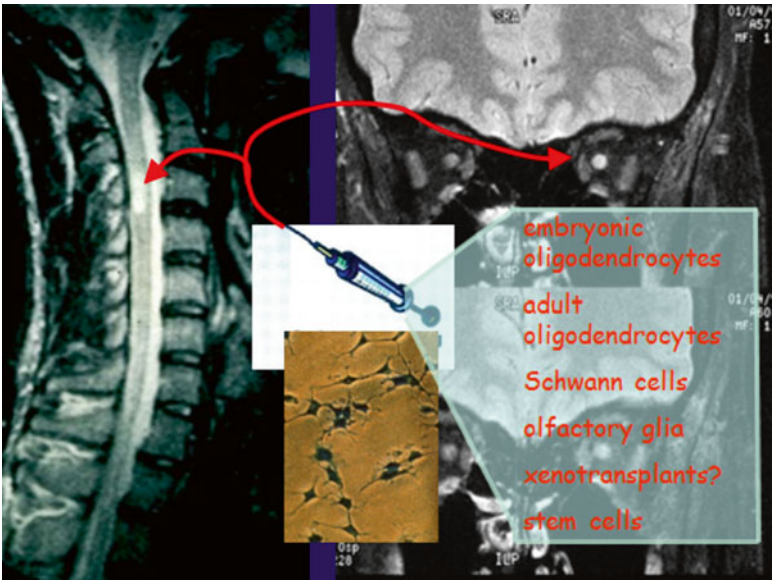

Fig. 14.1 The "classical" approach to cell therapy in MS—the direct injection of cells capable of producing myelin into MS lesions shown by MRI scanning

The initial enthusiasm for cell replacement was built on various aspects (as then understood) of MS that appeared to make this disease an ideal test bed for cell replacement therapy [7]. A single cell type was targeted by the immune system for damage, the oligodendrocyte; axons were preserved, so that remyelination should (and in rodent studies, could) restore efficient conduction and neurological function; demyelination occurred in focal patches that could readily be identified by MRI scanning and so putatively injected with remyelinating cells. And as mentioned above, rodent experiments appeared to prove the biological feasibility of this approach.

But a broader and deeper understanding of the biology of MS has cast considerable doubt on this rationale [4, 5]. Spontaneous remyelination in MS, first observed some 50 years ago and initially thought to be sparse and "abortive" [8], has been studied in great detail over the last decade in autopsy studies and found to be far more widespread and successful than hitherto thought [9, 10]—so that the underpinning need for promoting myelin repair has been questioned.

The explanation for this unexpectedly widespread spontaneous myelin repair is that endogenous oligodendrocyte progenitors are already present within MS lesions and present in significant numbers [11, 12]. There is also evidence that resident neural stem cells are likewise present and indeed that they react to disease processes by increasing their numbers [13]. The implication is that where remyelination does fail, the "problem" is more subtle than numbers of potential remyelinating cells: arrested maturation, disruption in the crucial initial interactions between axon and oligodendrocyte process, or other abnormalities of cell:cell signaling are postulated

explanations for the failure of myelin repair [14]. "Simply" adding exogenous cells appears unlikely to be the answer.

It might be asked why, if spontaneous myelin repair is so successful, the majority of MS patients develop significant disability? The answer further undermines the rationale of replacing myelinating cells as a therapy: namely that axon damage and also neuronal loss likely play a greater role than persistent demyelinated lesions in explaining progressive disability in MS [15, 16]. The disease is far from being one that exclusively damages oligodendrocytes.

And then finally, such neuro-axonal loss is now known to occur not only within focal (and MRI-disclosed) lesions, but diffusely, in gray and white matter throughout the brain and spinal cord. Focal injection of cells could do little for such diffuse damage.

That said, not all these "problems" necessarily always apply. MS also sometimes causes large demyelinating lesions which are likely to contribute to persistent disability [17]; and not all relapses are followed by complete recovery: spontaneous repair is not always sufficient. In these instances, focal injection of stem cells, aiming to supplement myelin repair, may well prove valuable. Additionally, inherited and other acquired disorders of myelin may involve lesions where permanent myelin loss is the principal cause of disability. Injecting stem cells as a source of remyelinating oligodendrocytes may still have promise in some clinical scenarios.

No less important, it is also undoubtedly true that the continuing study of remyelination biology, both stimulated and enabled by the prospect of cell therapy in MS, has proved remarkably successful in elucidating the cellular and molecular events underlying myelin repair [14]. Whilst "classical cell therapy"—injecting cells into lesions—has, as a consequence, become a somewhat less logical treatment strategy than formerly considered, insights from cell biology have offered a significant number of highly attractive molecular targets for small molecule or other drug-related therapeutic approaches specifically designed to promote and enhance spontaneous myelin repair [18, 19]. Delivered systemically, these would clearly offer the prospect of addressing more diffuse disease processes. The translation of such potential treatments from experimental to clinical studies in patients is advancing rapidly [20].

Immune Reconstitution: Hematopoietic Stem Cell Therapy

Autologous hematopoietic stem-cell transplantation (aHSCT) was originally conceived as an alternative to whole bone marrow transplantation, used to rescue patients from life-threatening bone marrow aplasia during the course of high-dose total body irradiation or myelo-ablative chemotherapy for leukemia. In multiple sclerosis (MS), a single dose of chemotherapy and/or total body irradiation is used with the aim of suppressing or preferably ablating the auto-destructive effector and memory cells of the immune system, allowing remission from MS autoimmune activity; aHSCT then offers the reconstitution of a normal (i.e., non-MS orientated)

immune and hematopoietic system [21]. This is fundamentally different from most conventional immune-modulatory or immunosuppressive regimens in aiming to restore tolerance and remove the autoimmune process, regenerating a fully functional immune system [22]. While there is little direct proof in treated MS patients that this does indeed occur, i.e., that autoimmune clones are eliminated [23], the clinical and radiological effects on inflammatory disease activity are substantial.

Impetus for the clinical translation of this approach was provided in the early 1990s by laboratory studies showing that high dose of cyclophosphamide or total body irradiation followed by syngenic bone marrow transplantation brought about complete inhibition of chronic relapsing autoimmune encephalomyelitis (CR-EAE) in the mouse, with a total inhibition of spontaneous relapses during a follow-up period of 2 months [24].

In patients, HSCs can be collected from the bone marrow by aspiration from the iliac crest or by drug-induced mobilization of peripheral blood HSC. Commonly used stem cell mobilization regimens include granulocyte-colony stimulating factor (G-CSF) administered concurrently with steroids and cyclophosphamide [21]. Prior to infusion, the collected graft can be manipulated to remove immune, auto-reactive T cells through the positive selection of $CD34^+$ cells or the negative deletion of T cells and frozen for storage while the patient undergoes "conditioning"—ablation or partial ablation of their immune system.

The most common conditioning regimen reported to European Group for Blood and Marrow Transplantation (EBMT) used to eradicate auto-reactive clones in the target organ is the BEAM regimen (carmustine, cytarabine, etoposide, and melphalan), all of which drugs can cross the blood–brain barrier [25]. Auto-reactive T cells can also be significantly depleted by the infusion of agents such as polyclonal antithymocyte globulin (ATG) or alemtuzumab. The intensity of the conditioning regimen must strike a balance between adequate immune ablation and regimen-related morbidity and mortality: safer, lower intensity regimens are increasingly explored. No single conditioning regimen has so far been shown markedly superiority to others [22]. Previous MS treatments such as interferons may affect aHSCT [22].

After completion of the conditioning regimen, the cryopreserved graft is thawed and the cells are infused. They then home to marrow space, where they seed and proliferate. Grafted cells mature into circulating blood cells and contribute to de novo lymphopoiesis.

The conditioning regimen is followed by the aplastic phase, but the graft allows recovery of the cell count some 10–20 days after infusion [26]. Expected effects from this include febrile neutropenia and infection. In the mobilizing and conditioning period, there may be relapses, and there is some suggestion that these may be associated with GCSF [26]. An engraftment syndrome consisting of noninfectious fever ± skin rash has occurred. Other late toxic effects (>100 days after transplantation) include Varicella Zoster infection and secondary autoimmune disorder (i.e., thyroiditis) [27].

The neurological outcome has been assessed in relatively small single center Phase I/II trials and in larger pooled studies [28–31]. The approach is effective in markedly reducing relapses. In relatively small studies (less than 75 patients), more

than 85 % of MS patients who received a conditioning regimen of BEAM and ATG were rendered free from clinical relapses in the absence of ongoing treatment with other disease-modifying agents [32, 33].

Some studies also reported beneficial effects on progression. For example, in one US open study of 21 patients, all were described as free from progression, and 16 were free of relapses, after a follow-up period of just over 3 years [34]. Some suggested, however, that more severely disabled patients, with a high pretransplantation disability score (EDSS > 6), were more likely to continue to deteriorate. Better outcomes were suggested for recipients younger than 40 years of age and diagnosed within the preceding 5 years [26].

It was also suggested that more "malignant" forms of MS, characterized by a rapidly evolving course with progression to severe disability, responded particularly well to aHSCT. In many reports of aHSCT in chronic MS, the reported improvement in EDSS scores was modest (0.5–1.0 range). By contrast, in patients with "malignant" MS, with follow-up extending to 4 years, EDSS scores dropped from a mean of 6.8 prior to aHSCT to a mean of 3.1 [35–37]. The inference was drawn that aHSCT might be more effective in the presence of active neuroinflammation [21].

MS patients undergoing aHSCT experienced comparable regimen-related complications to patients undergoing aHSCT for lymphoma. Urinary tract infections were common. MS patients with a greater degree of disability prior to transplantation were at risk of developing further loss of mobility due to chemotherapy-induced cachexia and myopathy. The risk of late opportunistic infections was small once immune reconstitution had occurred. Treatment-related mortality (TRM) when aHSCT was first introduced was much higher (up to 20 %) [38]. TRM during the period of 2000–2007 was reported to be significantly decreased to 1.3 % [27].

Clinical trials and observational studies continue. A recent single center experience of 123 relapsing-remitting and 28 secondary-progressive patients, with a median follow-up of 2 years, reported a 4-year relapse-free survival of 80 % and progression-free survival of 87 %. Importantly, post hoc analysis showed that disability (measured by EDSS) did not improve significantly in patients with secondary-progressive MS or in those with a disease duration of over 10 years [39]. Again, this would be consistent with the observed reduction in disability being at least partially explained by recovery from relapse. Indeed, one interpretation was that this study helped show that, while there was a clear, potent and lasting effect in suppressing relapses, "autologous HSCT does not appear to be effective against established progressive forms of MS and, absent new data, additional trials of these protocols are probably not indicated for patients with progressive MS" [23]. An additional complexity to interpreting these studies is that alemtuzumab was used in many conditioning regimens and does itself cause a substantial and sustained reduction in relapse rate. A further recent multicenter Phase II study, again including both relapsing-remitting and secondary progressive patients, also showed a substantial reduction in inflammatory disease activity and relapse frequency, but reported no effect on the progression of disability [40].

A joint EBMT and Centre for International Blood and Marrow Transplant Research (CIBMTR) registry-based, long-term follow-up study and a proposed

Phase III randomized trial of stem cell transplants versus best available therapy for patients with highly active MS who failed interferon-beta therapy will provide further information regarding outcome and benefit of aHSCT [41].

"Restorative" Cell Therapy

The classical properties of the stem cell are self-renewal and the ability to differentiate into multiple specialized cell types. It was these properties that first projected stem cells forward as a solution to the question of the ideal cell type to use to replace damaged oligodendrocytes in early cell therapy approaches to MS [7]. But it has become clear over the past decade or more that many stem cell types have additional potentially beneficial properties, unrelated to forming specialized cells. In some situations and with certain specific stem cell types, these "noncanonical" properties, some paracrine, others not, may play a considerably greater role in any therapeutic effect than conventional differentiation and cell replacement [42].

In relation to neurological disease, both neural stem cells and mesenchymal stem cells, the latter derived mainly from bone marrow (though in some studies also from other tissues, including adipose tissue) have been shown in experimental studies to have therapeutic potential that depends on such noncanonical properties [43–45]. Bone marrow-derived mesenchymal stem cells (Fig. 14.2)—which most authorities consider to have no capacity at all for differentiating into myelin-forming oligodendrocytes—have attracted probably the most attention.

Bone marrow has long been known to contain hematopoietic stem cells. But various other stem-cell types are also present, including mesenchymal stem cells (though there are others, and mesenchymal stem cells themselves are a heterogeneous population [46]). Furthermore, though first identified in bone marrow, mesenchymal stem cells are present in many tissues—indeed, in every tissue in which they

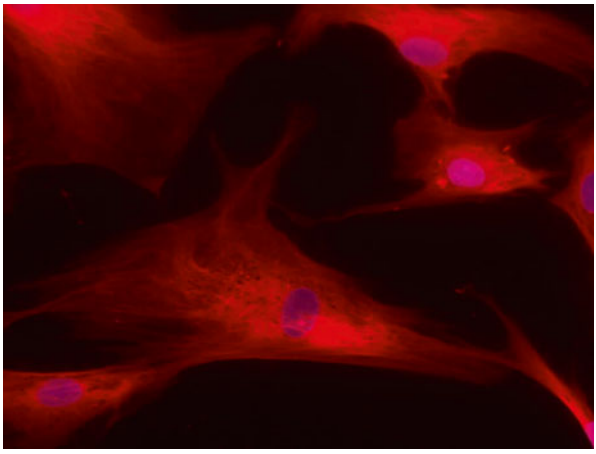

Fig. 14.2 Human mesenchymal cells growing in cell culture: cells with a wide range of potentially therapeutic properties [83]

have been sought [46, 47]. Their normal function within the bone marrow is to do with maintenance of the hematopoietic stem cell niche, but in addition there has been increasing evidence that mesenchymal stem cells have systemic activities to do with tissue repair. They may achieve such a function through multiple mechanisms, and many of these are relevant to MS and offer the prospect of ameliorating a number of various pathological processes now known collectively to contribute to the development of tissue damage in MS [42, 48]—what we have termed *restorative* cell therapy.

Remyelination

Studies commencing 15 years ago in experimental animals with nonimmune demyelination showed that not only isolated mesenchymal stem cells but also mixed populations of unseparated, nonexpanded bone marrow cells promote myelin repair following intravenous injection [49, 50]. The mode of action was not clear. Intravenously delivered bone marrow-derived cells successfully infiltrate the brain and spinal cord, inflamed or otherwise [51, 52], and they proliferate and migrate toward cytokines expressed in multiple sclerosis lesions [53]. Initially it was considered that bone marrow-derived cells arriving in demyelinated lesions might differentiate into Schwann cells and lay down peripheral-type myelin. Current thought, however, centers on the later-discovered ability of mesenchymal stem cells to interact with and stimulate local CNS endogenous neural precursors, encouraging both their proliferation [54], and their directed differentiation into oligodendrocytes [55]. Mesenchymal stem cells also secrete trophic factors for oligodendrocytes [56] which might additionally promote remyelination.

It is also reported that mesenchymal stem cells reduce gliotic scar formation in the CNS [57], gliosis being widely considered a major impediment to spontaneous myelin repair. They can also promote new blood vessel growth, and this too would also be expected to enhance tissue repair [58].

Suppressing Inflammation, Modulating Immunity

Bone marrow-derived cells have pronounced immune-modulating properties [59], affecting both innate and adaptive immune systems. Unsurprisingly, therefore, clinical effects in many systemic autoimmune diseases have been sought and in some cases benefit has been reported [60]. In relation to multiple sclerosis, numerous studies have shown both mesenchymal stem cells and mixed populations of bone marrow-derived cells successfully to abrogate various experimental allergic encephalomyelitis models through increasingly well-delineated immunosuppressive actions. Some authorities consider these immune effects sufficiently potent to justify clinical testing in relapsing-remitting MS [61] (MESEMS; *ClinicalTrials.gov Identifier* NCT01854957), almost irrespective of these cells' putative reparative or regenerative effects.

Neuroprotection

What of the progressive loss of axons and neurons in multiple sclerosis that contributes so greatly to the relentless accumulation of disability? Considering the enormous structural complexity of neuronal pathways, neuronal cell replacement approaches for brain repair remain extremely hard even to imagine for the foreseeable future. Reestablishing normal synaptic pathways in the developed CNS, capable of restoring function, appears a very remote prospect. The emphasis at present therefore remains on developing approaches to limit and reduce such damage and/or to ameliorate its consequences.

In multiple sclerosis, axon damage and neural cell loss likely result from several mechanisms. Inflammatory and immune mediators, possibly "sequestered" within the CNS, contribute [62], and so the immunomodulating/suppressing properties, both local and systemic, of bone marrow-derived cells are relevant and may potentially benefit. Mesenchymal stem cells reduce axon loss in various immune-mediated EAE models [63].

But they also help reduce axon damage in nonimmune CNS injury, including for example, experimental stroke models [64]. Here, other beneficial properties of these cells are more relevant. Human mesenchymal stem cells release superoxide dismutase-3 (SOD-3), with powerful neuroprotective effects [65]—and damage from reactive oxygen radicals is also postulated to occur in multiple sclerosis [66]. A range of neurotrophic factors, all constitutively synthesized, also contributes to these cells' neuroprotective properties. Mesenchymal stem cells also promote CNS neurite outgrowth and remodeling [67].

It is important to mention that adipose stromal cells likewise exhibit neuroprotective properties [68].

Cell Fusion

A particularly intriguing additional property of BMDCs has recently emerged, and this is cell fusion. Bone marrow-derived cells have long been known to fuse with certain differentiated cell types. The physiological significance of such fusion is, as yet, uncertain, but it appears quite clearly to occur in vivo as well as in vitro and can involve CNS neuronal cell types as "partner" cells [69]. Experimentally, local or systemic inflammation or immune activation promotes the fusion of circulating bone marrow cells with neurons following infiltration of the CNS, and this is seen with both rodent and also with human mesenchymal stem cells [70].

Fusion appears to represent a neuroprotective process by which healthy nuclei or functional genes from the mesenchymal stem cell are introduced into degenerating cells, helping to restore or rescue damaged neurons [71]. Rather extraordinarily, mesenchymal stem cells can also protect tissue by directly transferring mitochondria to vulnerable cells [72], membrane fusion (likely relating to nanotube formation or exosome transfer) representing the underlying mechanism common to both cell fusion and mitochondrial "donation." Preliminary evidence has emerged

that fusion of infiltrating (endogenous) bone marrow-derived cells with Purkinje cells, with subsequent heterokaryon formation, occurs spontaneously in MS patients [73].

Diffuse Damage

What of the question of multiple sclerosis as a nonlesional disease and the more diffuse gray matter disease and atrophy that form the key substrates of sustained disability in MS? Injecting cells into specific lesions could offer but little prospect of benefiting this aspect of the pathophysiology, but a cell therapy delivered systemically, rather like any conventional drug therapy, may well have more rationale—as well as being safer than a neurosurgical procedure.

Following intravenous injection, many cells are trapped in the lungs, but significant numbers still clearly enter the CNS and become widely distributed—not only in experimental models but in human subjects too [74], offering the clear possibility of a therapeutic effect where it is required. (Additionally, even cells "trapped" in the lungs may indirectly exert clinically relevant systemic anti-inflammatory therapeutic effects, clearly an intriguing area of future research.)

Others have explored delivery of bone marrow-derived cells in patients with neurological disease using injection into the carotid arteries (in multiple system atrophy, though not, as far as we are aware, in multiple sclerosis) [75]. Whilst appearing clinically safe, and while a higher proportion of injected cells would be expected to enter the CNS, there are indications that potentially hazardous microemboli form within the cerebral arterial system using this approach, which has constrained enthusiasm.

Clinical Translation

As with hematopoietic stem cell translation, restorative cell therapy using other bone marrow-derived cells delivered intravenously to exploit their reparative and neuroprotective effects has also begun the journey from laboratory to clinic—though only in more recent years so that published trials thus far are fewer and smaller. (The same is not necessarily the case in the clinical exploration of bone marrow-derived cells in other diseases: *ClinicalTrials.gov* currently lists around 2000 trials studying bone marrow-derived cells; and in myocardial infarction, both large scale Phase III randomized controlled trials and meta-analyses of trials are now reported.)

Various groups have published small safety and feasibility studies exploring autologous bone marrow-derived cell therapy in chronic multiple sclerosis, some using mixed/unseparated cells, others purified and expanded mesenchymal stem cells [76–80]. Most have utilized intravenous delivery, but intrathecal injection has

also been explored. The results have generally confirmed the safety and feasibility (though a transient meningeal syndrome is reported with intrathecal delivery); and some have reported preliminary and uncontrolled evidence from detailed neurophysiological studies of beneficial effects [77, 79]. Larger, controlled Phase II studies are now underway [81].

Conclusion

It is hopefully clear that, while the subject of stem cell therapy—or cell therapy—in multiple sclerosis has become increasingly complex and multifarious, this evolution has been a positive response to our rapidly advancing knowledge on the one hand of the underlying clinical biology of multiple sclerosis and on the other of stem cells and their various properties and types. It is hard to predict what this topic will look like in a decade or two. It has been suggested that many forms of cell therapy are no more than necessary stepping stones on a pathway that will rapidly see them replaced by more sophisticated forms of molecular therapy. We have already seen how the biological knowledge emerging from studies of classical oligodendrocyte replacement therapy have yielded therapeutic trials of small molecules designed to promote remyelination. Similarly, restorative cell therapy might, it is suggested, be successfully mimicked (obviating the need for cell harvest and preparation) by molecular therapies aiming to stimulate the release of specific cell populations from the bone marrow into the circulation, or by agents that enhance the migration of bone marrow derived cells from the circulation into neural tissue. What such approaches are unlikely, however, to reproduce, is what might be called the "afferent" side to cell therapy, a subject we have not touched upon. It is increasingly clear that the multiple potentially therapeutic capacities of which some bone marrow-derived populations are capable are not randomly activated in all disease situations: this is no "shot gun" effect. Rather, infiltrating cells sense and react: specific pathways are triggered in different tissues and in response to different forms of tissue damage and different disease processes [82]. It would be challenging to reproduce this by administering molecules rather than cells. There is as yet much mileage in the experimental and clinical exploration of cell therapy in relation to multiple sclerosis and indeed many other neurological diseases.

References

1. Blakemore WF. Remyelination of CNS axons by Schwann cells transplanted from the sciatic nerve. Nature. 1977;266(5597):68–9.
2. Duncan ID. Oligodendrocytes and stem cell transplantation: their potential in the treatment of leukoencephalopathies. J Inherit Metab Dis. 2005;28(3):357–68.
3. Stangel M, Hartung HP. Remyelinating strategies for the treatment of multiple sclerosis. Prog Neurobiol. 2002;68(5):361–76.

4. Lassmann H. Stem cell and progenitor cell transplantation in multiple sclerosis: the discrepancy between neurobiological attraction and clinical feasibility. J Neurol Sci. 2005;233(1–2):83–6. doi:10.1016/j.jns.2005.03.007.
5. Paty DW, Arnold DL. The lesions of multiple sclerosis. N Engl J Med. 2002;346(3):199–200.
6. Jose AM. Multiple sclerosis: can Schwann cells wrap it up? Yale J Biol Med. 2002;75(2):113–6.
7. Scolding NJ. Strategies for repair and remyelination in demyelinating diseases. Curr Opin Neurol. 1997;10:193–200.
8. Perier O, Gregoire A. Electron microscopic features of multiple sclerosis lesions. Brain. 1965; 88(5):937–52.
9. Albert M, Antel JP, Bruck W, Stadelmann C. Extensive cortical remyelination in patients with chronic multiple sclerosis. Brain Pathol. 2007;17(2):129–38.
10. Patani R, Balaratnam M, Vora A, Reynolds R. Remyelination can be extensive in multiple sclerosis despite a long disease course. Neuropathol Appl Neurobiol. 2007;33(3):277–87.
11. Maeda Y, Solanky M, Menonna J, Chapin J, Li W, Dowling P. Platelet-derived growth factor-alpha receptor-positive oligodendroglia are frequent in multiple sclerosis lesions. Ann Neurol. 2001;49(6):776–85.
12. Scolding NJ, Franklin RJM, Stevens S, Heldin CH, Compston DAS, Newcombe J. Oligodendrocyte progenitors are present in the normal adult human CNS and in the lesions of multiple sclerosis. Brain. 1998;121:2221–8.
13. Snethen H, Love S, Scolding N. Disease-responsive neural precursor cells are present in multiple sclerosis lesions. Regen Med. 2008;3(6):835–47.
14. Fancy SPJ, Kotter MR, Harrington EP, Huang JK, Zhao C, Rowitch DH, Franklin RJM. Overcoming remyelination failure in multiple sclerosis and other myelin disorders. Exp Neurol. 2010;225(1):18–23.
15. Chard D, Miller D. Is multiple sclerosis a generalized disease of the central nervous system? An MRI perspective. Curr Opin Neurol. 2009;22(3):214–8.
16. Evangelou N, DeLuca GC, Owens T, Esiri MM. Pathological study of spinal cord atrophy in multiple sclerosis suggests limited role of local lesions. Brain. 2005;128(Pt 1):29–34.
17. Hirst C, Ingram G, Pearson O, Pickersgill T, Scolding N, Robertson N. Contribution of relapses to disability in multiple sclerosis. J Neurol. 2008;255(2):280–7.
18. Najm FJ, Madhavan M, Zaremba A, Shick E, Karl RT, Factor DC, Tesar PJ. Drug-based modulation of endogenous stem cells promotes functional remyelination in vivo. Nature. 2015;522:216–20. doi:10.1038/nature14335.
19. Zhang Y, Zhang YP, Pepinsky B, Huang G, Shields LB, Shields CB, Mi S. Inhibition of LINGO-1 promotes functional recovery after experimental spinal cord demyelination. Exp Neurol. 2015;266:68–73. doi:10.1016/j.expneurol.2015.02.006.
20. Tran JQ, Rana J, Barkhof F, Melamed I, Gevorkyan H, Wattjes MP, Cadavid D. Randomized phase I trials of the safety/tolerability of anti-LINGO-1 monoclonal antibody BIIB033. Neurol Neuroimmunol Neuroinflamm. 2014;1(2), e18. doi:10.1212/NXI.0000000000000018.
21. Atkins HL, Freedman MS. Hematopoietic stem cell therapy for multiple sclerosis: top 10 lessons learned. Neurotherapeutics. 2013;10(1):68–76. doi:10.1007/s13311-012-0162-5.
22. Atkins H. Hematopoietic SCT for the treatment of multiple sclerosis. Bone Marrow Transplant. 2010;45(12):1671–81. doi:10.1038/bmt.2010.168.
23. Hauser SL. Hematopoietic stem cell transplantation for MS: extraordinary evidence still needed. JAMA. 2015;313(3):251–2. doi:10.1001/jama.2014.18150.
24. Karussis DM, Vourka-Karussis U, Lehmann D, Ovadia H, Mizrachi-Koll R, Ben-Nun A, Slavin S. Prevention and reversal of adoptively transferred, chronic relapsing experimental autoimmune encephalomyelitis with a single high dose cytoreductive treatment followed by syngeneic bone marrow transplantation. J Clin Invest. 1993;92(2):765–72. doi:10.1172/JCI116648.
25. Fassas A, Anagnostopoulos A, Kazis A, Kapinas K, Sakellari I, Kimiskidis V, Tsompanakou A. Peripheral blood stem cell transplantation in the treatment of progressive multiple sclerosis: first results of a pilot study. Bone Marrow Transplant. 1997;20(8):631–8. doi:10.1038/sj.bmt.1700944.

26. Saccardi R, Kozak T, Bocelli-Tyndall C, Fassas A, Kazis A, Havrdova E, Autoimmune Diseases Working Party of EBMT. Autologous stem cell transplantation for progressive multiple sclerosis: update of the European group for blood and marrow transplantation autoimmune diseases working party database. Mult Scler. 2006;12(6):814–23.
27. Mancardi G, Saccardi R. Autologous haematopoietic stem-cell transplantation in multiple sclerosis. Lancet Neurol. 2008;7(7):626–36. doi:10.1016/S1474-4422(08)70138-8.
28. Burt RK, Cohen BA, Russell E, Spero K, Joshi A, Oyama Y, Burns WH. Hematopoietic stem cell transplantation for progressive multiple sclerosis: failure of a total body irradiation-based conditioning regimen to prevent disease progression in patients with high disability scores. Blood. 2003;102(7):2373–8. doi:10.1182/blood-2003-03-0877.
29. Ni XS, Ouyang J, Zhu WH, Wang C, Chen B. Autologous hematopoietic stem cell transplantation for progressive multiple sclerosis: report of efficacy and safety at three yr of follow up in 21 patients. Clin Transplant. 2006;20(4):485–9. doi:10.1111/j.1399-0012.2006.00510.x.
30. Su L, Xu J, Ji BX, Wan SG, Lu CY, Dong HQ, Lu DP. Autologous peripheral blood stem cell transplantation for severe multiple sclerosis. Int J Hematol. 2006;84(3):276–81. doi:10.1532/IJH97.A10516.
31. Xu J, Ji BX, Su L, Dong HQ, Sun XJ, Liu CY. Clinical outcomes after autologous haematopoietic stem cell transplantation in patients with progressive multiple sclerosis. Chin Med J (Engl). 2006;119(22):1851–5.
32. Krasulová E, Trneny M, Kozák T, Vacková B, Pohlreich D, Kemlink D, Havrdová E. High-dose immunoablation with autologous haematopoietic stem cell transplantation in aggressive multiple sclerosis: a single centre 10-year experience. Mult Scler. 2010;16(6):685–93. doi:10.1177/1352458510364538.
33. Mancardi GL, Sormani MP, Di Gioia M, Vuolo L, Gualandi F, Amato MP, Italian BMT Study Group. Autologous haematopoietic stem cell transplantation with an intermediate intensity conditioning regimen in multiple sclerosis: the Italian multi-centre experience. Mult Scler. 2012;18(6):835–42. doi:10.1177/1352458511429320.
34. Burt RK, Loh Y, Cohen B, Stefoski D, Balabanov R, Katsamakis G, Burns WH. Autologous non-myeloablative haemopoietic stem cell transplantation in relapsing-remitting multiple sclerosis: a phase I/II study. Lancet Neurol. 2009;8(3):244–53.
35. Fagius J, Lundgren J, Oberg G. Early highly aggressive MS successfully treated by hematopoietic stem cell transplantation. Mult Scler. 2009;15(2):229–37. doi:10.1177/1352458508096875.
36. Kimiskidis V, Sakellari I, Tsimourtou V, Kapina V, Papagiannopoulos S, Kazis D, Fassas A. Autologous stem-cell transplantation in malignant multiple sclerosis: a case with a favorable long-term outcome. Mult Scler. 2008;14(2):278–83. doi:10.1177/1352458507082604.
37. Mancardi GL, Murialdo A, Rossi P, Gualandi F, Martino G, Marmont A, Uccelli A. Autologous stem cell transplantation as rescue therapy in malignant forms of multiple sclerosis. Mult Scler. 2005;11(3):367–71.
38. Openshaw H, Lund BT, Kashyap A, Atkinson R, Sniecinski I, Weiner LP, Forman S. Peripheral blood stem cell transplantation in multiple sclerosis with busulfan and cyclophosphamide conditioning: report of toxicity and immunological monitoring. Biol Blood Marrow Transplant. 2000;6(5A):563–75.
39. Burt RK, Balabanov R, Han X, Sharrack B, Morgan A, Quigley K, Burman J. Association of nonmyeloablative hematopoietic stem cell transplantation with neurological disability in patients with relapsing-remitting multiple sclerosis. JAMA. 2015;313(3):275–84. doi:10.1001/jama.2014.17986.
40. Mancardi GL, Sormani MP, Gualandi F, Saiz A, Carreras E, Merelli E, Marrow Transplantation E. Autologous hematopoietic stem cell transplantation in multiple sclerosis: a phase II trial. Neurology. 2015;84(10):981–8. doi:10.1212/WNL.0000000000001329.
41. Saccardi R, Freedman MS, Sormani MP, Atkins H, Farge D, Griffith LM, HSCT in MS International Study Group. A prospective, randomized, controlled trial of autologous haematopoietic stem cell transplantation for aggressive multiple sclerosis: a position paper. Mult Scler. 2012;18(6):825–34. doi:10.1177/1352458512438454.

42. Korbling M, Estrov Z. Adult stem cells for tissue repair. N Engl J Med. 2003;349:570–82.
43. Pluchino S, Zanotti L, Rossi B, Brambilla E, Ottoboni L, Salani G, Martino G. Neurosphere-derived multipotent precursors promote neuroprotection by an immunomodulatory mechanism. Nature. 2005;436(7048):266–71.
44. Rice CM, Kemp K, Wilkins A, Scolding NJ. Cell therapy for multiple sclerosis: an evolving concept with implications for other neurodegenerative diseases. Lancet. 2013;382(9899):1204–13. doi:10.1016/S0140-6736(13)61810-3.
45. Uccelli A, Moretta L, Pistoia V. Mesenchymal stem cells in health and disease. Nat Rev Immunol. 2008;8(9):726–36.
46. Phinney DG. Functional heterogeneity of mesenchymal stem cells: implications for cell therapy. J Cell Biochem. 2012;113(9):2806–12. doi:10.1002/jcb.24166.
47. da Silva ML, Chagastelles PC, Nardi NB. Mesenchymal stem cells reside in virtually all post-natal organs and tissues. J Cell Sci. 2006;119(Pt 11):2204–13.
48. Rice CM, Scolding NJ. Adult stem cells—reprogramming neurological repair? Lancet. 2004; 364(9429):193–9.
49. Akiyama Y, Radtke C, Honmou O, Kocsis JD. Remyelination of the spinal cord following intravenous delivery of bone marrow cells. Glia. 2002;39(3):229–36.
50. Sasaki M, Honmou O, Akiyama Y, Uede T, Hashi K, Kocsis JD. Transplantation of an acutely isolated bone marrow fraction repairs demyelinated adult rat spinal cord axons. Glia. 2001;35(1):26–34.
51. Devine SM, Cobbs C, Jennings M, Batholomew A, Hoffman R. Mesenchymal stem cells distribute to a wide range of tissues following systemic infusion into non-human primates. Blood. 2003;101:2999–3001.
52. Gordon D, Pavlovska G, Uney JB, Wraith DC, Scolding NJ. Human mesenchymal stem cells infiltrate the spinal cord, reduce demyelination, and localize to white matter lesions in experimental autoimmune encephalomyelitis. J Neuropathol Exp Neurol. 2010;69(11):1087–95.
53. Rice CM, Scolding N. Adult human mesenchymal cells proliferate and migrate in response to chemokines expressed in demyelination. Cell Adhes Migr. 2010;4:235–40.
54. Munoz JR, Stoutenger BR, Robinson AP, Spees JL, Prockop DJ. Human stem/progenitor cells from bone marrow promote neurogenesis of endogenous neural stem cells in the hippocampus of mice. Proc Natl Acad Sci U S A. 2005;102(50):18171–6.
55. Bai L, Lennon DP, Eaton V, Maier K, Caplan AI, Miller SD, Miller RH. Human bone marrow-derived mesenchymal stem cells induce Th2-polarized immune response and promote endogenous repair in animal models of multiple sclerosis. Glia. 2009;57:1192–203.
56. Pisati F, Bossolasco P, Meregalli M, Cova L, Belicchi M, Gavina M, Polli E. Induction of neurotrophin expression via human adult mesenchymal stem cells: implication for cell therapy in neurodegenerative diseases. Cell Transplant. 2007;16(1):41–55.
57. Li Y, Chen J, Zhang CL, Wang L, Lu D, Katakowski M, Chopp M. Gliosis and brain remodeling after treatment of stroke in rats with marrow stromal cells. Glia. 2005;49(3):407–17.
58. Bronckaers A, Hilkens P, Martens W, Gervois P, Ratajczak J, Struys T, Lambrichts I. Mesenchymal stem/stromal cells as a pharmacological and therapeutic approach to accelerate angiogenesis. Pharmacol Ther. 2014;143(2):181–96. doi:10.1016/j.pharmthera.2014.02.013.
59. Prockop DJ, Oh JY. Mesenchymal stem/stromal cells (MSCs): role as guardians of inflammation. Mol Ther. 2012;20(1):14–20.
60. Wang D, Zhang H, Liang J, Li X, Feng X, Wang H, Sun L. Allogeneic mesenchymal stem cell transplantation in severe and refractory systemic lupus erythematosus: 4 years experience. Cell Transplant. 2012;22:2267–2277.
61. Freedman MS, Bar-Or A, Atkins HL, Karussis D, Frassoni F, Lazarus H, Uccelli A. The therapeutic potential of mesenchymal stem cell transplantation as a treatment for multiple sclerosis: consensus report of the international MSCT study group. Mult Scler. 2010;16:503–10.
62. Mahad DH, Trapp BD, Lassmann H. Pathological mechanisms in progressive multiple sclerosis. Lancet Neurol. 2015;14(2):183–93. doi:10.1016/S1474-4422(14)70256-X.
63. Zhang J, Li Y, Lu M, Cui Y, Chen J, Noffsinger L, Chopp M. Bone marrow stromal cells reduce axonal loss in experimental autoimmune encephalomyelitis mice. J Neurosci Res. 2006;84(3): 587–95.

64. Chen J, Li Y, Wang L, Zhang Z, Lu D, Lu M, Chopp M. Therapeutic benefit of intravenous administration of bone marrow stromal cells after cerebral ischemia in rats. Stroke. 2001;32(4): 1005–11.
65. Kemp K, Hares K, Mallam E, Heesom KJ, Scolding N, Wilkins A. Mesenchymal stem cell-secreted superoxide dismutase promotes cerebellar neuronal survival. J Neurochem. 2009;114: 1569–80.
66. Witherick J, Wilkins A, Scolding N, Kemp K. Mechanisms of oxidative damage in multiple sclerosis and a cell therapy approach to treatment. Autoimmune Dis. 2010;2011:164608.
67. Shen LH, Xin H, Li Y, Zhang RL, Cui Y, Zhang L, Chopp M. Endogenous tissue plasminogen activator mediates bone marrow stromal cell-induced neurite remodeling after stroke in mice. Stroke. 2011;42(2):459–64.
68. Farinazzo A, Turano E, Marconi S, Bistaffa E, Bazzoli E, Bonetti B. Murine adipose-derived mesenchymal stromal cell vesicles: in vitro clues for neuroprotective and neuroregenerative approaches. Cytotherapy. 2015;17(5):571–8. doi:10.1016/j.jcyt.2015.01.005.
69. Johansson CB, Youssef S, Koleckar K, Holbrook C, Doyonnas R, Corbel SY, Blau HM. Extensive fusion of haematopoietic cells with Purkinje neurons in response to chronic inflammation. Nat Cell Biol. 2008;10(5):575–83.
70. Kemp K, Gordon D, Wraith DC, Mallam E, Hartfield E, Uney J, Scolding N. Fusion between human mesenchymal stem cells and rodent cerebellar Purkinje cells. Neuropathol Appl Neurobiol. 2010;37:166–78.
71. Kemp K, Wilkins A, Scolding N. Cell fusion in the brain: two cells forward, one cell back. Acta Neuropathol. 2014;128:629–38. doi:10.1007/s00401-014-1303-1.
72. Prockop DJ. Mitochondria to the rescue. Nat Med. 2012;18(5):653–4.
73. Kemp K, Gray E, Wilkins A, Scolding N. Purkinje cell fusion and binucleate heterokaryon formation in multiple sclerosis cerebellum. Brain. 2012;135(Pt 10):2962–72.
74. Cogle CR, Yachnis AT, Laywell ED, Zander DS, Wingard JR, Steindler DA, Scott EW. Bone marrow transdifferentiation in brain after transplantation: a retrospective study. Lancet. 2004;363(9419):1432–7.
75. Lee PH, Lee JE, Kim HS, Song SK, Lee HS, Nam HS, Sohn YH. A randomized trial of mesenchymal stem cells in multiple system atrophy. Ann Neurol. 2012;72(1):32–40.
76. Bonab MM, Sahraian MA, Aghsaie A, Karvigh SA, Hosseinian SM, Nikbin B, Gheini MR. Autologous mesenchymal stem cell therapy in progressive multiple sclerosis: an open label study. Curr Stem Cell Res Ther. 2012;7(6):407–14.
77. Connick P, Kolappan M, Crawley C, Webber DJ, Patani R, Michell AW, Chandran S. Autologous mesenchymal stem cells for the treatment of secondary progressive multiple sclerosis: an open-label phase 2a proof-of-concept study. Lancet Neurol. 2012;11(2):150–6.
78. Karussis D, Karageorgiou C, Vaknin-Dembinsky A, Gowda-Kurkalli B, Gomori JM, Kassis I, Slavin S. Safety and immunological effects of mesenchymal stem cell transplantation in patients with multiple sclerosis and amyotrophic lateral sclerosis. Arch Neurol. 2010;67(10):1187–94.
79. Rice CM, Mallam EA, Whone AL, Walsh P, Brooks DJ, Kane N, Scolding NJ. Safety and feasibility of autologous bone marrow cellular therapy in relapsing-progressive multiple sclerosis. Clin Pharmacol Ther. 2010;87(6):679–85.
80. Yamout B, Hourani R, Salti H, Barada W, El-Hajj T, Al-Kutoubi A, Kreidieh NM. Bone marrow mesenchymal stem cell transplantation in patients with multiple sclerosis: a pilot study. J Neuroimmunol. 2010;227:185–9.
81. Rice CM, Marks DI, Ben-Shlomo Y, Evangelou N, Morgan PS, Metcalfe C, Walsh P, Kane NM, Guttridge MG, Miflin G, Blackmore S, Sarkar P, Redondo J, Owen D, Cottrell DA, Wilkins A, Scolding NJ. Assessment of bone marrow-derived cellular therapy in progressive multiple sclerosis (ACTiMuS): study protocol for a randomised controlled trial. Trials. 2015 Oct 14;16:463. doi:10.1186/s13063-015-0953-1.
82. Murphy MB, Moncivais K, Caplan AI. Mesenchymal stem cells: environmentally responsive therapeutics for regenerative medicine. Exp Mol Med. 2013;45, e54. doi:10.1038/emm.2013.94.
83. Salewski RP, Mitchell RA, Li L, Shen C, Milekovskaia M, Nagy A, Fehlings MG. Transplantation of induced pluripotent stem cell-derived neural stem cells mediate functional recovery following thoracic spinal cord injury through remyelination of axons. Stem Cells Transl Med. 2015;4: 743–54. doi:10.5966/sctm.2014-0236.