Cell Proliferation

Cell Prolif., 2011, **44** (Suppl. 1), 35–38 doi: 10.1111/j.1365-2184.2010.00721.x

Adult stem cells and multiple sclerosis

N. Scolding

Department of Neurology, Frenchay Hospital, University of Bristol, Bristol, UK

Received 8 *June* 2010; *revision accepted* 19 *July* 2010

Abstract

Multiple sclerosis (MS) is a common neurological disease and a major cause of disability, particularly affecting young adults. It is characterized by patches of damage occurring throughout the brain and spinal cord, with loss of myelin sheaths – the insulating material around nerve fibres that allows normal conduction of nerve impulses – accompanied by loss of cells that make myelin (oligodendrocytes). In addition, we now know that there is damage to nerve cells (neurones) and their fibres (axons) too, and that this occurs both within these discrete patches and in tissue between them. The cause of MS remains unknown, but an autoimmune reaction against oligodendrocytes and myelin is generally assumed to play a major role, and early acute MS lesions almost invariably show prominent inflammation.

Efforts to develop cell therapy in MS have long been directed towards directly implanting cells capable of replacing lost oligodendrocytes and regenerating myelin sheaths. Accordingly, the advent of techniques to generate large numbers of oligodendrocytes from embryonic stem cells appeared a significant step towards new stem cell treatments for MS; while the emerging consensus that adult stem cells from, for example, the bone marrow had far less potential to turn into oligodendrocytes was thought to cast doubt on their potential value in this disease. A number of scientific and medical concerns, not least the risk of tumour formation associated with embryonic stem cells, have however, prevented any possible clinical testing of these cells in patients.

More recently, increasing understanding of the complexity of tissue damage in MS has emphasized that successful cell therapy may need to achieve far more than simply offering a source of replacement myelin-forming cells. The many and varied reparative properties of bone marrow-derived (mesenchymal) stem cells may well offer new and attractive possibilities for developing cell-based treatments for this difficult and disabling condition.

Correspondence: N. Scolding, Department of Neurology, Frenchay Hospital, University of Bristol, Bristol BS16 1LE, UK. Tel.: 44 1179 3406632; Fax: 44 117 975 3824; E-mail: n.j.scolding@bristol.ac.uk

Introduction

Multiple sclerosis (MS) is a common neurological disease, affecting approximately one in 800 people in most parts of Europe, and is a major cause of disability, particularly affecting young adults. It costs the European economy some €9 billion annually – not least through direct and indirect consequences of progressive disability in sufferers. MS has been known since its first description, to cause such disability, notwithstanding the defining (usual) relapsing–remitting initial course comprising discrete episodes of neurological dysfunction, sub-acute in onset and resolving spontaneously, and often completely, over a period of weeks. The great majority of patients experience such relapses, but in addition, over 80% of MS patients also develop *progressive* disease, where disability accumulates very slowly – over years rather than weeks or months – but inexorably. Forty per cent of patients require a wheelchair within 10 years of diagnosis.

It is, at present, incurable. We do not know the cause of the disease – a complex interplay of environmental and genetic factors culminating in autoimmune attack within the brain and spinal cord (central nervous system, CNS) is generally the accepted synthesis, but what such environmental factors may be remains obscure. Immune treatments are therefore routinely used, and these can reduce individual relapses both in severity (steroids, given acutely) and in frequency (interferons, glatiramer, and more recently various monoclonal antibodies, taken regularly). However, immune treatments have no impact on patients with progressive disability – their neurological deficits continue relentlessly, to accumulate.

Our therapeutic impotence in this neurodegenerative disorder has stimulated much searching for alternative approaches to treatment – as with many other currently incurable neurological diseases, such as Parkinson's and Alzheimer's diseases. One striking possibility that has steadily gained in credence over the past 30 years or so has been that of cell therapy – using exogenous cells to repair CNS damage in MS.

In this brief review, I hope to show why conventional strategies for applying cell therapy, using stem cells, to multiple sclerosis may need revising to accommodate recent developments in our understanding of the pathophysiology of the disease, which present serious barriers to orthodox stem cell approaches to MS. But I hope also to show that in parallel, advances in our knowledge of certain stem cell populations, particularly of bone marrow cells, can be seen to meet and match these challenges, and that such adult cells offer genuine prospects in this and other neurodegenerative diseases. Finally I will summarize our own efforts in Bristol, building on our and others' laboratory and experimental studies, to begin clinical translation of bone marrow cell therapy, describing our small phase one clinical trial studying autologous bone marrow cell infusion in six patients with chronic MS.

Possibility of repair in multiple sclerosis

Autoimmune processes characterizing multiple sclerosis appear to be directed against oligodendrocytes, cells in the CNS responsible for synthesizing and maintaining myelin. Myelin is the 'insulating' material that is wrapped around axons and so facilitates conduction of nerve impulses. Both oligodendrocytes and myelin are lost within patches of inflammation in MS – so-called lesions or plaques, which are readily seen on MRI scans and which, broadly, correspond to individual clinical relapse.

The nature of this process appeared to lend itself extremely well to some form of reparative cell therapy. As the autoimmune process appeared so targeted and specific, and only a single cell type, the oligodendrocyte required to be replaced. Axons appeared largely spared, and so the insurmountably complex challenge of re-building circuitry did not apply. Also, the ability to demonstrate the site of lesions using MRI scans would allow cells to be injected accurately into the midst of the lesion – where, the hope was, they would generate new myelin restoring normal impulse conduction and with that, improved neurological function.

Some 30 years ago, Bill Blakemore showed in a remarkable study that exogenous myelinating cells (Schwann cells in this instance), injected into a demyelinated lesion in the rodent CNS, could indeed achieve successful remyelination (1). This and subsequent studies offered vital proof of principle for remyelinating cell therapy for patients with MS (2). A key remaining problem was to identify a source of cells for injection into patients, and emergence of embryonic stem cells and their potential oligodendrocyte progeny were thought to offer a clear potential solution.

Problems of therapeutic repair in multiple sclerosis

There are, however, some problems with this theoretical approach, and these are listed below:

- It was shown in the 1960s that, perhaps surprisingly, a certain amount of spontaneous myelin repair occurred in MS (3). Initially, this was considered both partial, and sparse, but recent studies have shown that it is far more widespread, and successful, than previously thought (4,5); this raises the question of whether, as spontaneous myelin repair is so successful, attempts to augment the process are as pressingly needed as first thought.
- More recently, studies of MS lesions have shown that oligodendrocyte progenitors, the cells responsible for spontaneous myelin repair, are present in lesions in significant numbers (6–10) as indeed are endogenous neural precursors (11). So this raises the question of whether adding still further stem cells or oligodendrocytes would actually help.
- Furthermore, although lesions are the pathophysiological basis for relapses, it is increasingly considered that chronic disability in MS is more closely correlated with far more diffuse neuronal and axonal damage in the brain and spinal cord, rather than lesions (12–15). Repairing discrete lesions might therefore not usefully affect chronic progressive disability, and axon and neuronal damage, rather than oligodendrocyte loss, appeared to be the key problems in progressive MS.

Possibilities of bone marrow cells

If straightforward oligodendrocyte replacement into lesions to make new myelin is, after all, not a reasonable approach, how might cell therapy make progress in MS?

We (and many others) have concentrated on bone marrow cells partly because of the evidence that normal function of adult stem cells is (spontaneous) tissue repair (16), and that they achieve this through multiple mechanisms (17) – many of which are particularly apposite to a disease such as MS. As it happens, it may well be the case that among these properties, transdifferentiation, at least into oligodendrocytes, is not possible for the much-studied mesenchymal stem cell sub-population of bone

marrow cells. However, as outlined above, cell therapy in MS may well no longer have as its core aim replacing myelinating cells to regenerate myelin, and as shown below, indirect means of promoting remyelination are nonetheless apparent.

These cells do, however, exhibit a number of other functional capacities that are tantalizingly attractive in the context of considering what might be useful in MS:

- They have pronounced immune-modulating properties of significant possible relevance to MS (18–21).
- Both whole, unseparated, bone marrow cells, and isolated MSCs, can promote myelin repair after experimental (non-immune) demyelination (22,23). Mechanisms remain uncertain, but indirect effects seem to be more likely – bone marrow cells can stimulate proliferation of endogenous neural stem cells (24) and direct their differentiation along oligodendrocyte pathways (25–27).
- Intravenously delivered cells successfully infiltrate the brain and spinal cord, and are stimulated to migrate towards those cytokines known to be expressed in MS lesions, so that local actions are likely to be important (28).
- MSCs also appear to reduce gliotic scar formation – gliosis representing a major barrier to spontaneous repair (26,29).
- MSCs also secrete a wide range of neuroprotective factors, including both growth factors such as NGF, BDNF and GDNF (among others) (30–34) and antioxidants such as superoxide dismutase 3 (35).
- Fusion of MSCs with some cell types (perhaps particularly cerebellar Purkinje cells) may represent a further mechanism by which MSCs may offer protection (or rescue damaged cells) (36,37).
- Finally, ability of intravenously delivered cells to infiltrate throughout the human CNS, and apparently remain in place for years if not decades (38,39), offers further encouragement, particularly given the likelihood that injecting cells directly into lesions is unlikely to be useful.

Time to begin translation

At some point, modelling and theorizing must yield to translation into clinical use. Such a point is marked by a significant body of experimental evidence suggesting potential clinical benefit, and a much larger evidence base for suspecting safety. Given the studies and considerations outlined above, taken in context of a wealth of historical evidence attesting to safety of bone marrow cells, from haematological medicine over many decades, and from our more adventurous cardiological (and other) colleagues in recent years (40,41), we elected to commence translational investigations with a small safety and feasibility study exploring bone marrow cell therapy in chronic MS.

In close collaboration with our haematology colleagues, we treated six patients in an open label study of autologous cells, harvesting bone marrow in each under general anaesthetic, filtering and re-suspending cells, and re-injecting an average of 9×10^{10} cells intravenously into each patient. The procedure was safe and well-tolerated, with secondary outcome measures providing preliminary evidence on extensive neurophysiological testing of beneficial effects (42).

Stem cell therapy for neurological disease

While plainly there is much to do, and we are at the earliest possible stage of developing these therapies, results so far are encouraging. Significantly, range of non-canonical repair mechanisms mentioned above could well be relevant to neurological and neurodegenerative diseases other than multiple sclerosis. In Parkinson's disease, motor neuron disease, Alzheimer's disease and other disorders, neurons are lost, oxidative damage is often implicated as a final common pathway, and immune and inflammatory responses, even if secondary rather than primary, play a role. The ability of MSCs to offer neuroprotection, secrete growth factors and antioxidants, and to be attracted to areas of damage within the CNS after intravenous delivery, their autologous nature, and relative accessibility, all offer promise. Re-focusing (stem) cell therapy on the more general – if complex and varied – processes comprising tissue repair, and exploiting promiscuously reparative and non-canonical properties of (for example) bone marrow cells rather than over-concentrating on replacement of individual cell populations, may usefully accelerate translation in some diseases.

References

1 Blakemore WF (1977) Remyelination of CNS axons by Schwann cells transplanted from the sciatic nerve. *Nature* **266**, 68–69.
2 Scolding NJ (1997) Strategies for repair and remyelination in demyelinating diseases. *Curr. Opin. Neurol.* **10**, 193–200.
3 Perier O, Gregoire A (1965) Electron microscopic features of multiple sclerosis lesions. *Brain* **88**, 937–952.
4 Albert M, Antel J, Bruck W, Stadelmann C (2007) Extensive cortical remyelination in patients with chronic multiple sclerosis. *Brain Pathol.* **17**, 129–138.
5 Patani R, Balaratnam M, Vora A, Reynolds R (2007) Remyelination can be extensive in multiple sclerosis despite a long disease course. *Neuropathol. Appl. Neurobiol.* **33**, 277–287.
6 Chang A, Nishiyama A, Peterson J, Prineas J, Trapp BD (2000) NG2-positive oligodendrocyte progenitor cells in adult human brain and multiple sclerosis lesions. *J. Neurosci.* **20**, 6404–6412.

7 Maeda Y, Solanky M, Menonna J, Chapin J, Li W, Dowling P (2001) Platelet-derived growth factor-alpha receptor-positive oligodendroglia are frequent in multiple sclerosis lesions. *Ann. Neurol.* **49**, 776–785.

8 Scolding N, Franklin R, Stevens S, Heldin CH, Compston A, Newcombe J (1998) Oligodendrocyte progenitors are present in the normal adult human CNS and in the lesions of multiple sclerosis. *Brain* **121**, 2221–2228.

9 Wilson HC, Scolding NJ, Raine CS (2006) Co-expression of PDGF alpha receptor and NG2 by oligodendrocyte precursors in human CNS and multiple sclerosis lesions. *J. Neuroimmunol.* **176**, 162–173.

10 Wolswijk G (1998) Chronic stage multiple sclerosis lesions contain a relatively quiescent population of oligodendrocyte precursor cells. *J. Neurosci.* **18**, 601–609.

11 Snethen H, Love S, Scolding N (2008) Disease-responsive neural precursor cells are present in multiple sclerosis lesions. *Regen. Med.* **3**, 835–847.

12 Chard D, Miller D (2009) Is multiple sclerosis a generalized disease of the central nervous system? An MRI perspective. *Curr. Opin. Neurol* **22**, 214–218.

13 Evangelou N, DeLuca GC, Owens T, Esiri MM (2005) Pathological study of spinal cord atrophy in multiple sclerosis suggests limited role of local lesions. *Brain* **128**, 29–34.

14 Filippi M, Rocca MA (2005) MRI evidence for multiple sclerosis as a diffuse disease of the central nervous system. *J. Neurol.* **252**(Suppl. 5), v16–v24.

15 Kutzelnigg A, Lucchinetti CF, Stadelmann C, Bruck W, Rauschka H, Bergmann M *et al.* (2005) Cortical demyelination and diffuse white matter injury in multiple sclerosis. *Brain* **128**, 2705–2712.

16 Korbling M, Estrov Z (2003) Adult stem cells for tissue repair. *N. Engl. J. Med.* **349**, 570–582.

17 Rice CM, Scolding NJ (2004) Adult stem cells – reprogramming neurological repair? *Lancet* **364**, 193–199.

18 Gerdoni E, Gallo B, Casazza S, Musio S, Bonanni I, Pedemonte E *et al.* (2007) Mesenchymal stem cells effectively modulate pathogenic immune response in experimental autoimmune encephalomyelitis. *Ann. Neurol.* **61**, 219–227.

19 Gordon D, Pavlovska G, Glover CP, Uney JB, Wraith D, Scolding NJ (2008) Human mesenchymal stem cells abrogate experimental allergic encephalomyelitis after intraperitoneal injection and with sparse CNS infiltration. *Neurosci. Lett.* **448**, 71–73.

20 Zappia E, Casazza S, Pedemonte E, Benvenuto F, Bonanni I, Gerdoni E *et al.* (2005) Mesenchymal stem cells ameliorate experimental autoimmune encephalomyelitis inducing T-cell anergy. *Blood* **106**, 1755–1761.

21 Zhang J, Li Y, Chen J, Cui Y, Lu M, Elias SB *et al.* (2005) Human bone marrow stromal cell treatment improves neurological functional recovery in EAE mice. *Exp. Neurol.* **195**, 16–26.

22 Akiyama Y, Radtke C, Honmou O, Kocsis JD (2002) Remyelination of the spinal cord following intravenous delivery of bone marrow cells. *Glia* **39**, 229–236.

23 Akiyama Y, Radtke C, Kocsis JD (2002) Remyelination of the rat spinal cord by transplantation of identified bone marrow stromal cells. *J. Neurosci.* **22**, 6623–6630.

24 Munoz JR, Stoutenger BR, Robinson AP, Spees JL, Prockop DJ (2005) Human stem/progenitor cells from bone marrow promote neurogenesis of endogenous neural stem cells in the hippocampus of mice. *Proc. Natl. Acad. Sci. USA* **102**, 18171–18176.

25 Bai L, Caplan A, Lennon D, Miller RH (2007) Human mesenchymal stem cells signals regulate neural stem cell fate. *Neurochem. Res.* **32**, 353–362.

26 Bai L, Lennon DP, Eaton V, Maier K, Caplan AI, Miller SD *et al.* (2009) Human bone marrow-derived mesenchymal stem cells induce Th2-polarized immune response and promote endogenous repair in animal models of multiple sclerosis. *Glia* **57**, 1192–1203.

27 Rivera FJ, Couillard-Despres S, Pedre X, Ploetz S, Caioni M, Lois C *et al.* (2006) Mesenchymal stem cells instruct oligodendrogenic fate decision on adult neural stem cells. *Stem Cells* **24**, 2209–2219.

28 Rice CM, Scolding N (2010) Adult human mesenchymal cells proliferate and migrate in response to chemokines expressed in demyelination. *Cell Adh. Migr.* **4**(2), 235–40.

29 Li Y, Chen J, Zhang CL, Wang L, Lu D, Katakowski M *et al.* (2005) Gliosis and brain remodeling after treatment of stroke in rats with marrow stromal cells. *Glia* **49**, 407–417.

30 Chen Q, Long Y, Yuan X, Zou L, Sun J, Chen S *et al.* (2005) Protective effects of bone marrow stromal cell transplantation in injured rodent brain: synthesis of neurotrophic factors. *J. Neurosci. Res.* **80**, 611–619.

31 Garcia R, Aguiar J, Alberti E, de la Cuetara K, Pavon N (2004) Bone marrow stromal cells produce nerve growth factor and glial cell line-derived neurotrophic factors. *Biochem. Biophys. Res. Commun.* **316**, 753–754.

32 Li Y, Chen J, Chen XG, Wang L, Gautam SC, Xu YX *et al.* (2002) Human marrow stromal cell therapy for stroke in rat: neurotrophins and functional recovery. *Neurology* **59**, 514–523.

33 Wilkins A, Kemp K, Ginty M, Hares K, Mallam E, Scolding N (2009) Human bone marrow-derived mesenchymal stem cells secrete brain-derived neurotrophic factor which promotes neuronal survival in vitro. *Stem Cell Res.* **3**, 63–70.

34 Ye M, Chen S, Wang X, Qi C, Lu G, Liang L *et al.* (2005) Glial cell line-derived neurotrophic factor in bone marrow stromal cells of rat. *NeuroReport* **16**, 581–584.

35 Kemp K, Hares K, Mallam E, Heesom KJ, Scolding N, Wilkins A (2009) Mesenchymal stem cell-secreted superoxide dismutase promotes cerebellar neuronal survival. *J. Neurochem.* **114**, 1569–1580.

36 Johansson CB, Youssef S, Koleckar K, Holbrook C, Doyonnas R, Corbel SY *et al.* (2008) Extensive fusion of haematopoietic cells with Purkinje neurons in response to chronic inflammation. *Nat. Cell Biol.* **10**, 575–583.

37 Nygren JM, Liuba K, Breitbach M, Stott S, Thoren L, Roell W *et al.* (2008) Myeloid and lymphoid contribution to non-haematopoietic lineages through irradiation-induced heterotypic cell fusion. *Nat. Cell Biol.* **10**, 584–592.

38 Cogle CR, Yachnis AT, Laywell ED, Zander DS, Wingard JR, Steindler DA *et al.* (2004) Bone marrow transdifferentiation in brain after transplantation: a retrospective study. *Lancet* **363**, 1432–1437.

39 Mezey E, Key S, Vogelsang G, Szalayova I, Lange GD, Crain B (2003) Transplanted bone marrow generates new neurons in human brains. *Proc. Natl. Acad. Sci. USA* **100**, 1364–1369.

40 Burt RK, Loh Y, Pearce W, Beohar N, Barr WG, Craig R *et al.* (2008) Clinical applications of blood-derived and marrow-derived stem cells for nonmalignant diseases. *JAMA* **299**, 925–936.

41 Rice CM, Scolding NJ (2008) Autologous bone marrow stem cells – properties and advantages. *J. Neurol. Sci.* **15**, 59–62.

42 Rice CM, Mallam EA, Whone AL, Walsh P, Brooks DJ, Kane N *et al.* (2010) Safety and feasibility of autologous bone marrow cellular therapy in relapsing-progressive multiple sclerosis. *Clin. Pharmacol. Ther.* **87**, 679–685.