Adult bone marrow: which stem cells for cellular therapy protocols in neurodegenerative disorders?

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ABSTRACT

The generation of neuronal cells from stem cells obtained from adult bone marrow is of significant clinical interest in order to design new cell therapy protocols for several neurological disorders. The recent identification in adult bone marrow of stem cells derived from the neural crests (NCSC) might explain the neuronal phenotypic plasticity shown by bone marrow cells. However, little information is available about the nature of these cells compared to mesenchymal stem cells (MSC). In this manuscript, we will review all information available concerning NCSC from adult tissues and their possible use in regenerative medecine. Moreover, as multiple recent studies showed the beneficial effect of bone marrow stromal cells in neurodegenerative diseases, we will discuss which stem cells isolated from adult bone marrow should be more suitable for cell replacement therapy.

INTRODUCTION

Neurodegenerative disease is a generic term used for a wide range of acute and chronic conditions whose etiology is unknown such as Parkinson's disease, Huntington's disease, amyotrophic lateral sclerosis (ALS), Alzheimer's disease, but also now for other neurological diseases whose etiology is better known but which are also concerned by a chronic lost of neurons and glial cells such as multiple sclerosis (MS), stroke, and spinal cord injury. Although the adult brain contains small numbers of stem cells in restricted areas, the central nervous system exhibits limited capacity of regenerating lost tissue. Therefore, cell replacement therapies of lesioned brain have provided the basis for the development of potentially powerful new therapeutic strategies for a broad spectrum of human neurological diseases. However, the paucity of suitable cell types for cell replacement therapy in patients suffering from neurological disorders has hampered the development of this promising therapeutic approach.

Stem cells are classically defined as cells that have the ability to renew themselves continuously and possess pluripotent or multipotent ability to differentiate into many cell types. Besides the germ stem cells devoted to give rise to ovocytes or spermatozoids, those cells can be classified in three subgroups: embryonic stem cells (ES), induced pluripotent stem cells (iPS) and somatic stem cells (Figure 1). ES cells are derived from the inner mass of blastocyst and are considered as pluripotent stem cells as these cells can give rise to various mature cells from the three germ layers. iPS cells are also pluripotent stem cells, however, those cells derived from adult somatic cells such as skin fibroblasts are genetically modified by introduction of four embryogenesis-related genes (Takahashi et al., 2007; Park et al., 2008). Finally, tissue-specific stem cells known as somatic or adult stem cells are more restricted stem cells (multipotent stem cells) and are isolated from various fetal or adult tissues (i.e. hematopoietic stem cells, bone marrow mesenchymal stem cells, adipose tissue-derived stem cells, amniotic fluid stem cells, neural stem cells, etc.; Reviewed by Kim and de Vellis, 2009).

In recent years, neurons and glial cells have been successfully generated from stem cells such as embryonic stem cells (Patani et al., 2010), iPS (Swistowski et al., 2010), mesenchymal stem cells (MSC) (Wislet-Gendebien et al., 2005), and adult neural stem cells (reviewed by Ming et Song, 2011), and extensive efforts by investigators to develop stem cell-based brain transplantation therapies have been carried out. Over the last decade, convincing evidence has emerged of the capability of various stem cell populations to induce regeneration in animal models of Parkinson’s disease (PD), Huntington’s disease, Alzheimer’s disease (AD), multiple sclerosis or cerebral ischemia (Reviewed by Gögel et al., 2011). Some of the studies have already been carried out to clinical trials. In example, in the case of Parkinson’s disease, transplantation of fetal ventral mesencephalon tissue directly into the brains of PD patients has been done in a few centers with varying results (Kordower et al., 2008; Li et al., 2008 ; Mendez et al., 2008) and it appeared that using fetal ventral mesencephalon tissue raised numerous problems from ethical issues to heterogeneity and relative scarcity of tissue (reviewed by Wakeman et al., 2011) suggesting that other stem cells (like adult somatic stem cells) may be more suitable for such a therapy. Likewise, ES cells have also been grafted in patients with injured spinal cord, as USA Federal Regulators have cleared the way for the first human trials of human ES-cell research, authorizing researchers to test whether those cells are safe or not (Schwarz et al., 2010). It is still to early to know the effect of ES cells on patient recovery; however, several concerns have been previously raised on animal models as ES cells induced

teratocarcimas and some exploratory clinical trials are confirming the animal studies (reviewed by Solter, 2006).

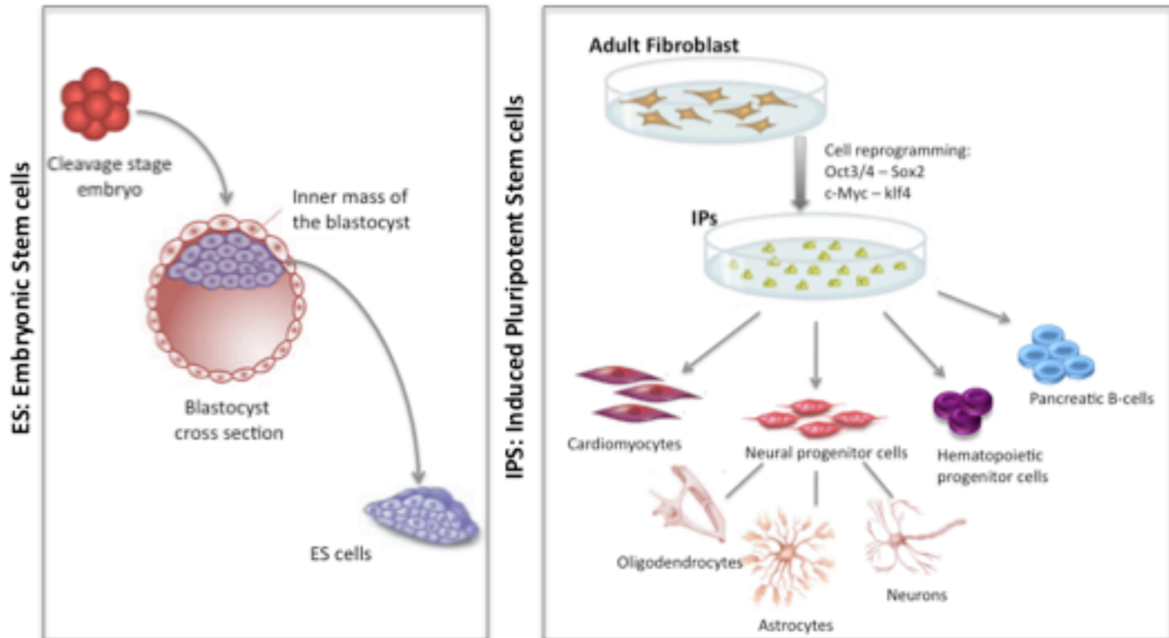
In this chapter, we will review our results concerning identification and characterization of neural crest stem cells (NCSC) in adult bone marrow as a potential source for cellular therapy in neurological disorders. We will also discuss what are the main questions that remain pending concerning the use of those cells in cellular therapy protocols for neurological disorders.

SOMATIC STEM CELLS ISOLATED FROM ADULT BONE MARROW

The post-natal bone marrow has traditionally been seen as an organ composed of two main systems rooted in distinct lineages—the hematopoietic tissue and the associated supporting stroma. The evidence pointing to a putative stem cell upstream of the diverse lineages and cell phenotypes comprising the bone marrow stromal system has made marrow the only known organ in which two (or more) separate and distinct stem cells and dependent tissue systems not only coexist but functionally cooperate, defining hematopoietic stem cells (HSC) and mesenchymal stem cells (MSC) (reviewed by Bianco et al., 2001).

MSC were first isolated from the bone marrow (BM-MSC) stem cell niche. More recently, extensive research has revealed that cells with morphological and functional characteristics similar to BM-MSC can be identified in a large number of organs or tissues including adipose tissue and peripheral blood. Despite having different origins, these MSC populations maintain cell biological properties typically associated with stem cells. These include continuous cell cycle progression for self-renewal and the potential to differentiate into highly specialized cell types of the mesodermal phenotype including chondroblast, osteoblast, and adipocyte lineages. Interestingly, BM-MSC have also been reported to be inducible via the ectodermal or endodermal germline, demonstrating the expression of neuron-like factors insulin production or hepatic lineage-associated genes respectively. In addition to these general stem cell properties, the International Society for Cellular Therapy proposed a more specific panel of markers for the characterization of MSC. Due to the failure to identify a certain unique MSC cell-surface molecule, a set of minimal criteria for MSC was recommended, which includes the capability of adherence to plastic surfaces and the expression of the cell surface markers CD44, CD73, CD90, and CD105 with a concomitant absence of CD14, CD19, CD34, CD45, and HLA-DR expression (Reviewed by Hilfiker et al., 2011).

Originally analyzed because of their critical role in the formation of the hematopoietic microenvironment (HME), bone marrow stromal cells became interesting because of their surprising ability to differentiate into mature neural cell types. More recently, a third stem cell group has been identified as originating from the neural crest, which could explain the capacity of stromal stem cells to differentiate into functional neurons.



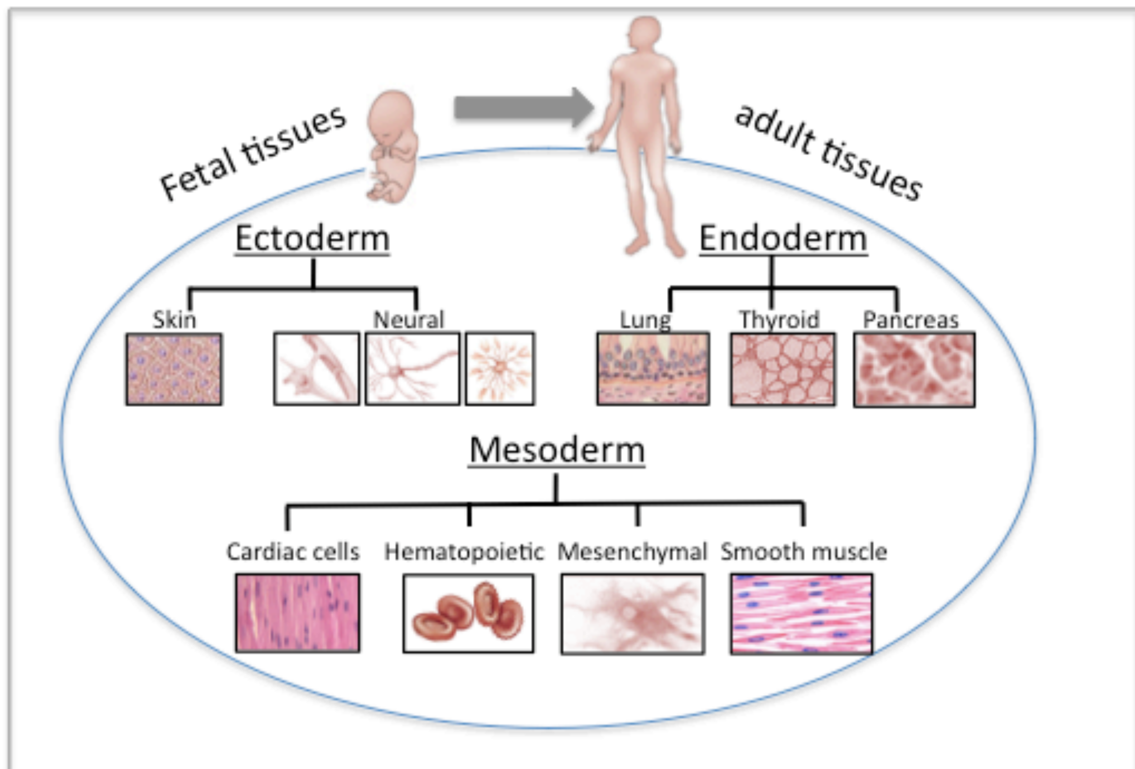


Figure 1. Stem cell type and origin. Besides germ stem cells, three group of stem cells can be defined according to their differentiating abilities: **A.** pluripotent embryonic stem cells (ES), **B.** induced pluripotent stem cells (iPS) and **C.** multipotent fetal or adult somatic stem cells.

NEURAL PHENOTYPIC PLASTICITY OF ADULT BONE MARROW STROMAL CELLS.

Several years ago, we demonstrated that a fraction of bone marrow stromal cells were able to differentiate into functional neurons. Those specific cells were characterized as nestin-positive mesenchymal stem cells (Wislet-Gendebien, 2003-2005). Electrophysiological analyses using the whole-cell patch-clamp technique revealed that adult rat bone marrow stromal cells (Wislet-Gendebien et al., 2005a and 2005b) were able to differentiate into excitable neuron-like cells when they were co-cultivated with mouse cerebellar granule neurons. First, we demonstrated that those cells express several neuronal markers (NeuN and Beta-III-tubulin ; Figure 2), an axonal marker (neurofilament H and M protein recognized by the monoclonal antibody, SMI31) and a dendritic marker (MAP2ab). Electrophysiological recordings of these nestin-positive bone marrow-derived neuron-like cells (BMDN) were performed and three maturation stages were observed (Table 1).

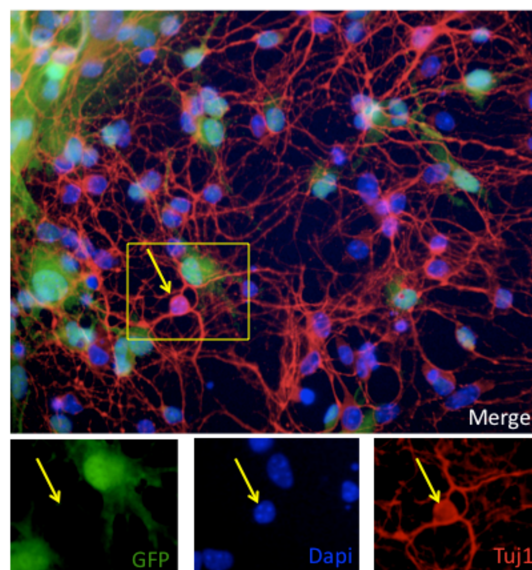


Figure 2. Neuronal marker expressed by bone marrow stromal cells. Bone marrow stromal cells were co-cultivated for 5 days with GFP-positive cerebellar granule neurons (green). Immunofluorescence labeling showed that beta-III tubulin recognize by Tuj1 antibodies (red) was expressed by about 20% of bone marrow stromal cells (GFP-negative or non-green cells) (Wislet-Gendebien et al., 2005).

At 4–6 days of co-culture, BMDN showed some neurotransmitter responsiveness (GABA, glycine, serotonin and glutamate) and voltage-gated K^+ currents inhibited by TEA (tetraethylammonium). However, those cells did not express functional sodium voltage-gated channels and have a high membrane potential (*V*rest) (-37.6° ± 3mV, *n* = 61). During the second week of co-culture, BMDN started to display Na^+ currents reversely inhibited by TTX (tetrodotoxin) and became able to fire single spike of action potential. In those older co-cultures, the *V*rest reaches a more negative value, which was closer to the value usually measured in neurons (7–9 days, -50.3 ± 2mV, *n* = 76 and 10–15 days, -56.7 ± 2.3mV, *n* = 97).

Maturation of BMDN	5 Days in vitro	8 Days in vitro	12 Days in vitro
Neurotransmitter sensitivities	GABA, Glycin, Glutamate	GABA, Glycin, Glutamate	GABA, Glycin, Glutamate
Potassic voltage-gated channels	+++	+++	+++
Sodic voltage-gated channels	_	+++	+++
Action potentials	_	+++	+++
Trains of action potentials	_	_	_
Synaptic activities	_	_	_
Membrane potential (mV)	-37 ± 3	-50,3 ± 2	- 57,7 ± 2,3

Table 1. Maturation steps of bone marrow derived neuron-like cells

As only nestin-positive bone marrow stromal cells were able to differentiate into functional neurons, we performed several proteomic and transcriptomic comparisons that pointed out several characteristics like ErbB3 and Sox10 over-expression in nestin-positive MSC, suggesting that these cells could actually be neural-crest derived cells (reviewed by Wislet-Gendebien et al., 2008). Few months later, Nogoshi et al. (2008) confirmed the presence of neural crest derived cells in adult bone marrow.

CHARACTERIZATION OF NEURAL CREST STEM CELLS FROM ADULT BONE MARROW

Neural crest stem cell origin

In early vertebrate development, the neural crest is specified in the embryonic ectoderm at the boundary of the neural plate and the ectoderm. Once specified, the neural crest cells undergo a process of epithelium to mesenchyme transition (EMT) that will confer them the ability to migrate. The EMT involves different molecular and cellular machineries and implies deep changes in cell morphology and in the type of cell surface adhesion and recognition molecules. When the EMT is complete, they delaminate from the neural folds/neural tube and migrate along characteristic pathways to differentiate into a wide variety of derivates (**Figure 3**; reviewed by Kalcheim, 2000).

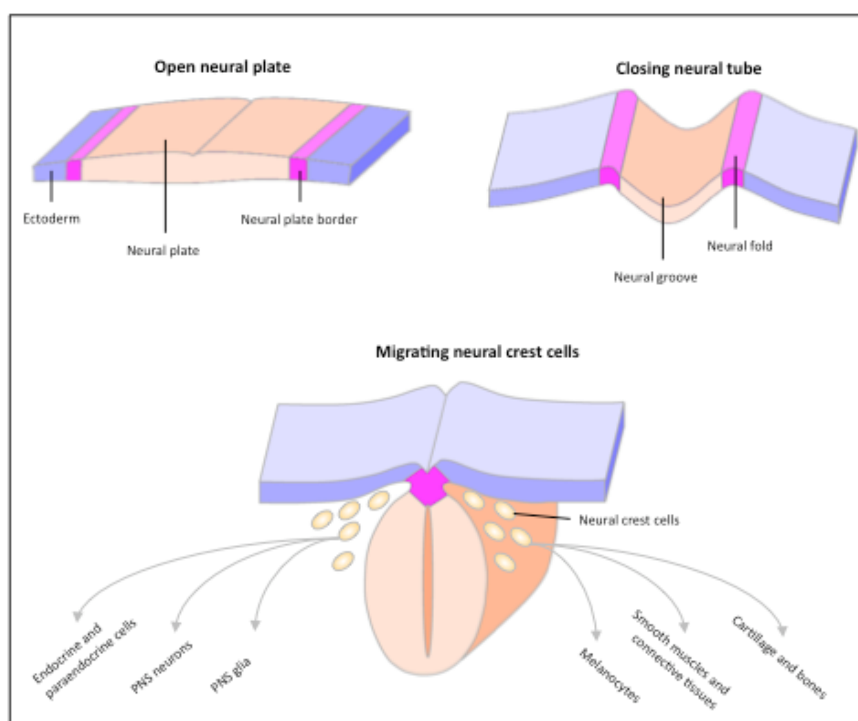


Figure 3. Neurulation and neural crest migration. As neurulation proceeds, the neural plate rolls up and the neural plate border becomes the neural folds. Near the time of neural tube closure (depending on the species), the neural crest cells go through an epithelial to mesenchymal transition (EMT) and delaminate from the neural folds or dorsal neural tube and migrate along defined pathways.

Takashima et al. (2007) was the first to address the biological origin of MSC and showed that they are generated in waves, with the neuroepithelium unexpectedly providing the first wave and a second wave of nonneuralderived MSC taking precedence in the adult (Reviewed by Miller, 2007). Indeed, using protocols that differentiate ES cells to mesodermal versus neural/neural crest lineages, they demonstrated that both lineages generated PDGFRa-positive cells (a marker for MSC) that could make adipocytes. However, the surprise came when they found that the neural, but not mesodermal, differentiations contained MSC that could proliferate extensively as multipotent clones. Moreover, these MSC were generated from cells expressing Sox1, a definitive marker for neuroepithelium, demonstrating their neural origin. Thus, for ES cells, différentiation along a mesodermal pathway did not generate MSCs, but différentiation toward a neural/neural crest fate did.

In order to address the *in vivo* relevance of these findings Takashima et al. (2007) used a transgenic mice expressing GFP under Sox1 promoter. They then isolated the trunk of these embryos at E9.5 (thereby excluding the cranial neural crest, which is known to generate mesenchymal cells) and demonstrated that Sox1-GFP-positive cells gave rise to PDGFRa-positive MSC. In contrast, GFP-negative, PDGFRa-positive cells (which expressed mesodermal markers) did not generate MSC, although they did make adipocytes. Thus, just as seen with ES cells, MSC could be generated from trunk neuroepithelial cells, but not from mesodermal cells in mid-gestation embryos. These experiments demonstrated that trunk neuroepithelium could make MSC. To demonstrate that it actually did so, the authors made Sox1-Cre/YFP mice in which the progeny of Sox1-positive neuroepithelial cells were persistently labeled and confirmed the presence of YFP cells in adult bone marrow.

Place	Marker	Animal	Genotype	Reference
Gut	P75NTR	Rat	Wild type	Kruger et al., 2002
DRG		Rat	Wild type	Li et al., 2007
DRG, Whisker pad, bone marrow	EGFP	Mouse	P0 Wnt1-CRE/CAG-EGFP	Nagoshi et al., 2008
Skin		Mouse	Wild type	Toma et al., 2005
Skin	LacZ	Mouse	Wnt1-CRE/ROSA-LacZ	Sieber-Blum et al., 2004
Skin	EYFP	Mouse	Dct-Cre/ROSA-EYFP	Wong et al., 2006
Cornea	EGFP	Mouse	P0 Wnt1-CRE/CAG-EGFP	Yoshida et al., 2006
Carotide body	EGFP	Mouse	GFAP promotor-EGFP	Pardal et al., 2007

Table 2. Presence of neural crest derived cells in adult tissues.

In parallel, using a two-component genetic system based on Cre/lox recombination to label indelibly the entire mouse neural crest population at the time of its formation (Jiang et al., 2000), several groups used *Wnt1-Cre/R26R* double transgenic mice, in which virtually all neural crest stem cells express β-galactosidase, to identified NCSC in various tissues. Indeed, using this transgenic model, Sieber-Blum and Grim (2004) demonstrated the presence of pluripotent neural crest stem cells in adult follicle hairs, Wong et al. (2006) demonstrated the presence of neural crest cells in the mouse adult skin and Nagoshi et al. (2008) confirmed the presence of NCSC in adult bone marrow (Table 2).

Self-renewal ability and multipotency of adult bone marrow NCSC

To consider NCSC from adult bone marrow as a potential source for cellular therapy protocol, a better characterization of those cells was mandatory. In our study, we first address the self-renewal ability, as first characteristic of stemness. Indeed, we demonstrated that NCSC were able to grow as spheres, which is one of the main hallmarks of immature neural cells and proliferate from a single cell culture (clonal culture). We then addressed the multipotency and verify if those NCSC clones were able to differentiate into multiple mature cell types. Indeed, we observed that NCSC were able to differentiate into adipocytes, melanocytes, smooth muscles, osteocytes, neurons and astrocytes (Figure 4, Glejzer et al., 2011).

Maintenance and proliferation of adult bone marrow NCSC.

Before using NCSC from adult bone marrow, we have to face some limiting factors like the fact that NCSC are a minority population (less than 1%) in adult bone marrow. As Wnt1 and BMP2 factors were described to help for maintenance and proliferation of NCSC isolated from embryo (Sommer, 2006), we tested those two factors, on adult NCSC isolated from adult bone marrow. Interestingly, we demonstrated that Wnt1 and BMP2 were able to increase the number of NCSC present in bone marrow stromal cell culture, up to four times within 2 passages (Glejzer et al., 2011) reaching 20 % of NCSC.

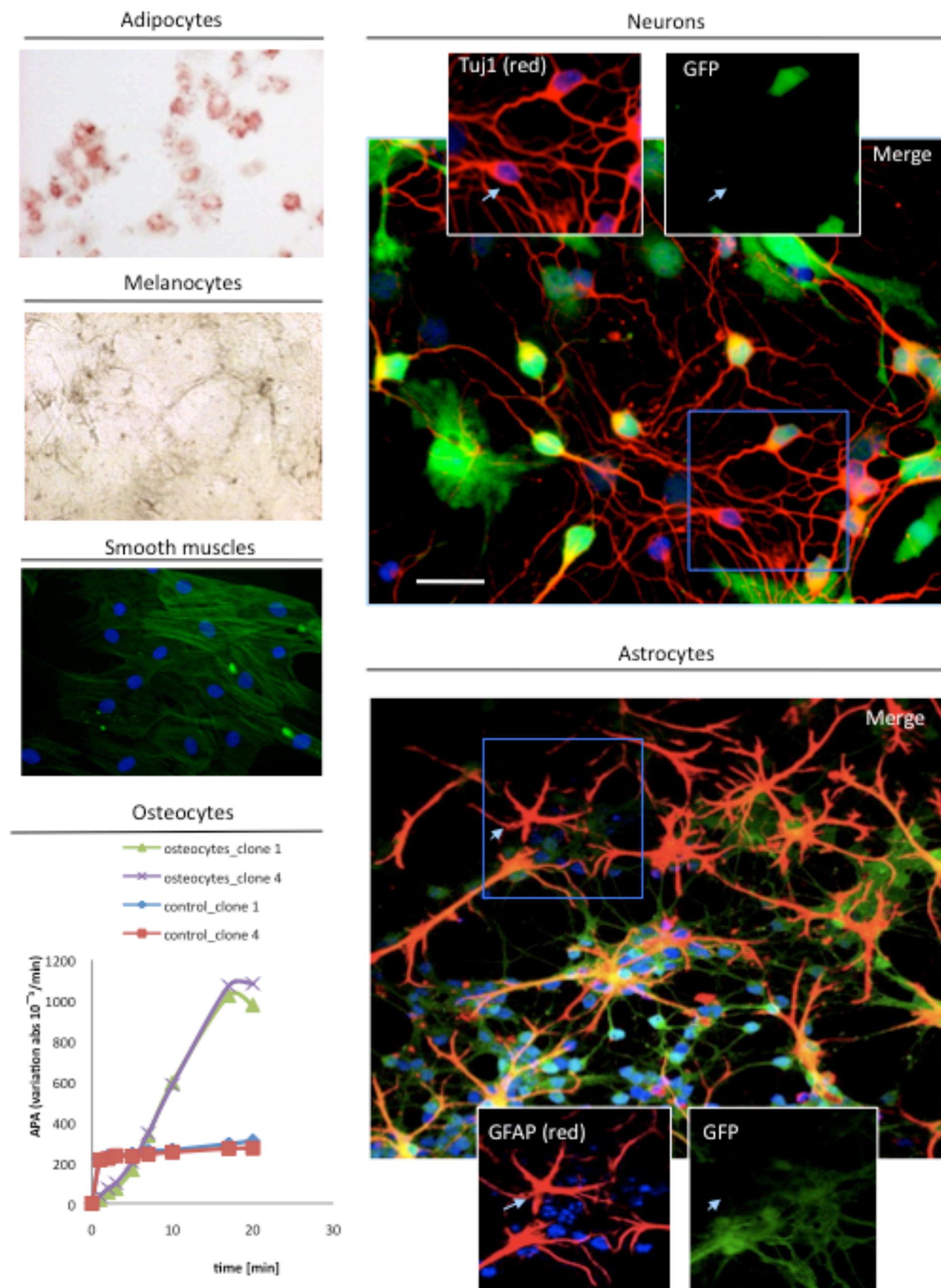


Figure 4. Multipotency of adult bone marrow NCSC. NCSC clones were subjected to differentiating protocols and were shown to be able to differentiate into adipocytes (Oil Red O labeling), melanocytes (L-DOPA labeling), smooth muscles (SMA-labeling) and Osteocytes (alkaline phosphatase activity). Moreover, when co-cultured with cerebellar granule neurons, we were able to differentiate NCSC clones into neurons (betaIII-tubulin labeling by Tuj1 monoclonal antibody) or glial cells (GFAP labeling).

IN VIVO **CHARACTERIZATION OF NEURAL CREST STEM CELLS AND/OR BONE MARROW STROMAL CELLS IN NEUROLOGICAL DISORDER MICE MODELS.**

Spinal stroke

Among others, the spinal cord is the collection of fibers that runs from or to the brain through the spine, carrying signals from or to the brain to or from the rest of the body. Those signals control a person's muscles and enable the person to feel various sensations. The main consequence of injuries to the spinal cord is the interference with those signals. Those injuries are characterized as "complete" or "incomplete": if the injured person loses all sensation and all ability to control the muscles below the point of the injury, the injury is said "complete"; in the case of an "incomplete" injury, the victim retains some ability to feel sensations or control movement below the injured area.

Main goals in spinal cord repair include reconnecting brain and lower spinal cord, building new circuits, re-myelination of demyelinated axons, providing trophic support, and bridging the gap of the lesion (Reviewed by Enzmann et al., 2006). Overcoming myelin-associated and/or glial-scar-associated growth inhibition are experimental approaches that have been most successfully studied in *in vivo* experiments. Further issues concern gray matter reconstitution and protecting neurons and glia from secondary death (Reviewed by Enzmann et al., 2006).

In this purpose, neural crest stem cells isolated from the bulge of hair follicle have been grafted in rat model of spinal cord lesion (reviewed by Sieber-Blum 2010). Those cells survived, integrated and intermingled with host neurites in the lesioned spinal cord. NCSC were non-migratory and did not proliferate or form tumors. Significant subsets of grafted cells expressed the neuron-specific beta-III tubulin, the GABAergic marker glutamate decarboxylase 67 (GAD67), the oligodendrocyte markers RIP or myelin basic protein (MBP) (Sieber-Blum et al., 2006). More interestingly, functional improvement was shown by two independent approaches, spinal somatosensory evoked potentials (SpSEP) and the Semmes-Weinstein touch test (Hu et al., 2010). The strength of NSCS was fully characterized as they can exert a combination of pertinent functions in the contused spinal cord, including cell replacement, neuroprotection, angiogenesis and modulation of scar formation. However, those results have never been confirmed with human NCSC, which should be the next promising step.

Similar studies were previously performed with bone marrow stromal cells. Indeed, several researches reported the anti-proliferative, anti-inflammatory and anti-apoptotic features of bone marrow stromal cells (reviewed by Uccelli et al., 2011). Indeed, Zeng et al. (2011) demonstrated that BMSC seeded in a three dimensions gelatin sponge scaffold and transplanted in a transected rat spinal cord resulted in attenuation of inflammation, promotion of angiogenesis and reduction of cavity formation. Those BMSC were isolated from 10 weeks old rats and passaged 3 to 6 times. Likewise, Xu et al. (2010) demonstrated that a co-culture of Schwann cell with BMSC had greater effects on injured spinal cord recovery than untreated BMSC. Indeed, analyses of chemokine and cytokine expression revealed that BMSC/Schwann cell co-cultures produced far less MCP-1 and IL-6 than BMSC or Schwann cells cultured alone. Transplanted BMSC may thus improve recovery in spinal cord injured mice through immunosuppressive effects that can be enhanced by a Schwann cell co-culturing step. These results indicate that the temporary presence of BMSC in the injured cord is

sufficient to alter the cascade of pathological events that normally occurs after spinal cord injury and therefore, generating a microenvironment which favours an improved recovery. In this study, BMSC were isolated from adult mice and used after 4 passages.

Krabbe's disease

Krabbe's disease, a demyelinating disorder caused by mutations in the lysosomal enzyme galactocerebrosidase (GALC), is a disorder of the nervous system where cell transplantation is the only available therapy (Oliveira Miranda et al., 2011). In this leukodystrophy, apoptosis of myelin-forming oligodendrocytes and Schwann cells is caused by accumulation of a GALC substrate, galactosylsphingosine (psychosine), which causes a severe demyelination of both the peripheral (PNS) and central nervous system (CNS). Effective treatment of Krabbe's disease is challenging given the rapid decline of patients and the need to correct both the PNS and CNS.

So far, the most effective treatment for Krabbe's patients is hematopoietic stem cell (HSC) transplantation, which supplies the missing enzyme to the nervous system, however, this option showed only a mild and temporary beneficial effect on peripheral nerves. As a consequence of a lack of appropriate treatment, a recent study analyzed the therapeutical properties of MSC in such a disease (Oliveira Miranda et al., 2011). The authors demonstrated that MSC had a multi-level mechanism of action targeting neurons, Schwann cells and macrophages that co-ordinately promoted recovery of nerve pathology following intravenous transplantation, demonstrating that MSC could also be used in peripheral nervous system pathology.

Multiple sclerosis

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 accompanied by loss of cells that make myelin (oligodendrocytes) (reviewed by Scolding, 2011). In addition, we now know that there is damage to neurons and their 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 of nervous system lesion in MS have long been directed towards directly implanting cells capable of replacing lost oligodendrocytes and regenerating myelin sheaths.

To our knowledge, no experiment has been performed to characterize the effect of neural crest stem cells on the improvement of Multiple Sclerosis disease; however, several data can be collected concerning the positive effect of Schwann cells (derived from NCSC) and of bone marrow stromal cells.

As previously described in injured spinal cord, bone marrow stromal cells have been characterized on their anti-proliferative, anti-inflammatory and anti-apoptotic features. These properties have been exploited in the effective treatment of experimental autoimmune encephalomyelitis (EAE), an animal model of multiple sclerosis where the inhibition of the autoimmune response resulted in a significant neuroprotection (reviewed by Uccelli et al., 2011). Based on recent experimental data, a number of clinical trials have been designed for

the intravenous (IV) and/or intrathecal (ITH) administration of BMSCs in MS patients (Grigoriadis et al., 2011).

Parkinson disease

Parkinson's disease (PD) is a chronic, progressive neurodegenerative disorder characterized by a continuous and selective loss of dopaminergic neurons in the *substantia nigra pars compacta* with a subsequent reduction of dopamine release mainly in the striatum. This ongoing loss of nigral dopaminergic neurons leads to clinical diagnosis mainly due to occurrence of motor symptoms such as rigidity, tremor and bradykinesia, which result from a reduction of about 70% of striatal dopamine (reviewed by Meyer et al., 2010).

Levy et al. (2008) analyzed the effect of differentiated human BMSC onto dopaminergic precursor on hemi-Parkinsonian rats, after transplantation into striatum. This graft resulted in improvement of rat behavioral deficits quantified by apomorphine-induced rotational behavior. The transplanted induced-neuronal cells proved to be of superior benefit compared with the transplantation of naive BMSC. Immunohistochemical analysis of grafted brains revealed that abundant induced cells survived the grafting procedure and some of these cells displayed dopaminergic traits.

Similarly, Zhang et al. (2008) isolated and characterized MSCs from **Parkinson**'s **disease** (PD) patients and compared them with MSCs derived from normal adult **bone marrow**. These authors show that PD-derived MSCs are similar to normal MSCs in phenotype, morphology, and differentiation capacity. Moreover, PD-derived MSCs are able of differentiating into neurons in a specific medium with up to 30% having the characteristics of dopamine cells. At last, PD-derived MSCs could inhibit T-lymphocyte proliferation induced by mitogens. These findings indicate that MSCs derived from PD patients' **bone marrow** could be a promising cell type for cellular therapy and somatic gene therapy applications.

Huntington disease

Huntington disease (HD) is an autosomal dominant genetic disorder caused by the expansion of polyglutamine encoded by CAG repeats in Exon 1 of the *IT15* gene encoding for Huntingtin (Htt). The polyglutamine repeat length determines the age of onset and the overall level of function, but not the severity of the disease (Vassos et al., 2007). Although the exact mechanism underlying HD disease progression remains uncertain, the hallmark of this disease is a gross atrophy of the striatum and cortex and a decrease of GABAergic neurons (DiFiglia et al., 1997).

One strategy for HD therapy is to enhance neurogenesis, which has been studied by the administration of Stem/progenitor cells, including BMSC. Several studies (reviewed by Snyder et al., 2010) showed that BMSC promote repair of the CNS by creating a more favorable environment for neuroprotection and regeneration through the secretion of various cytokines and chemokines. Moreover, Snyder et al. (2010) demonstrated that BMSC injected into the dentate gyrus of HD mice model increased neurogenesis and decreased atrophy of the striatum.

Alzheimer disease

Alzheimer's disease (AD) is the most common form of dementia, affecting more than 18 million people worldwide. With increased life expectancy, this number is expected to rise in the future. AD is characterized by progressive memory deficits, cognitive impairment, and personality changes associated with the degeneration of multiple neuronal types and pathologically by the presence of neuritic or amyloid plaques and neurofibrillary tangles (Reviewed by Selko, 2001). Amyloid β-peptide (Aβ) appears to play a key pathogenic role in AD, and studies have connected Aβ plaques with the formation of intercellular tau tangles, another neurotoxic feature of AD (Reviewed by Mattson, 2004). Currently, no treatment is available to cure or prevent the neuronal cell death that results in inevitable decline in AD patients.

The innate immune system is the vital first line of defense against a wide range of pathogens and tissue injuries, triggering inflammation through activation of microglia and macrophages. Many studies have shown that microglia are attracted to and surround senile plaques both in human AD samples and in rodent transgenic models that develop AD-related disease (Simard et al., 2006). In this context, Lee and al. (2010) demonstrated that treated APP/PS1 mice (mouse model of AD) with BM-MSCs promoted microglial activation, rescued cognitive impairment, and reduced Aβ and tau pathology in the mouse brain.

CONCLUSIONS

The NCSC is one of the most intriguing cells in the field of regenerative medicine, because it is easily harvested from various accessible peripheral tissues, which could make autologous transplantation possible. Autologous transplantation would avoid immunological complications as well as the ethical concerns associated with the use of embryonic stem cells. Of the various NCSC, research on skin-derived NCSC is the most advanced mainly due to their easy isolation process. One of the critical questions for the application of NCSC to regenerative medicine is whether cells that are differentiated from NCSCs are functional. Some evidence supports this (reviewed by Nagoshi et al., 2009), however, lots of questions remained pending. By example, a very important question is the differentiation abilities of NCSC isolated from various tissues: are they similar or different?

On the other hand, even if bone marrow stromal cells did not show a strong ability to replace lost neurons in neurodegenerative disorders such as Parkinson or Huntington disease, their impact on inflammation modulation or stimulation of endogenous cells were quite remarkable. This impact is also illustrated by a high number of ongoing clinical trials with these cells (Reviewed by Sensebé et Bourin, 2011). However, the main challenges remain the standardization of cell culture and isolation, to meet the international rules. Indeed, more than ever, it has been demonstrated that bone marrow stromal cells are constituted of an heterogenous population containing multiple stem/progenitor cell types including mesenchymal stem cells and neural crest stem cells, among other. Most of the studies describing the effects of BMSC on inflammation modulation or stimulation of endogenous cells were performed on low passages (< 4), which mainly contain MSC and less than 10 % of NCSC. So we could stipulate that most of these effects were probably due to MSC. However, in a perspective of cell therapy, a strong characterization of the role of each cell type in neuronal recovery seemed mandatory to establish strong and safe protocols.

ACKNOWLEDGMENT

Our work presented in this review was supported by grants from the Fonds National de la Recherche Scientifique (FNRS) of Belgium, by a grant of the Action de Recherche Concertée de la Communauté Française de Belgique, and by the Belgian League against Multiple Sclerosis associated with Leon Fredericq Foundation.

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