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REVIEW ARTICLE

HEMATOPOIETIC GROWTH FACTORS: NOVEL THERAPEUTIC STRATEGY FOR ALZHEIMER'S DISEASE

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ABSTRACT

The search for trophic factors that impact the degenerating nervous system has expanded beyond the classical neurotrophins. There is increasing recognition that hematopoietic growth factors (HGFs) have neurotrophic properties. This review will focus specifically on HGFs or cytokines that have direct and indirect effects on the brain and which may have therapeutic potential for neurodegenerative diseases, including Alzheimer's disease (AD). These include stem cell factor (SCF), granulocyte colony-stimulating factor (G-CSF), monocyte colony-stimulating factor (M-CSF), granulocyte–macrophage colony-stimulating factor (GM-CSF) and erythropoietin (EPO). HGFs have the capacity to prevent neuronal death, stimulate the proliferation and differentiation of neural stem/progenitor cells (NSCs) into new neurons, enhance the formation of new synapses, modulate inflammatory processes, mobilize monocytes from the peripheral circulation and increase their infiltration into the brain, where they reinforce the endogenous microglial population. G-CSF has been safely and routinely administered to enhance hematopoiesis in bone marrow donors and is now being tested for therapeutic efficacy and safety in patients with stroke and AD. HGF receptors expressed on neurons and NSCs provide novel targets for drug discovery in the search for agents that can forestall or reverse the pathological progression of AD.

INTRODUCTION

The most common cause of dementia in the elderly is Alzheimer's disease (AD), characterized pathologically by accumulation of extracellular β-amyloid plaques, neurofibrillary tangles and synaptic loss in regions of the brain important for learning and memory (1). Other characteristics include diminished neuronal metabolism and disturbances of multiple transmitter systems (2, 3). The prominent changes in the cholinergic system of the basal forebrain logically led to the development of pharmacotherapy with acetylcholinesterase inhibitors (AChE-I). These agents may transiently improve memory but have no significant effects on the progressive degenerative process. However, the observation that cholinergic neurons degenerate in the absence of nerve growth factor (NGF) led to numerous studies of the neuroprotective effects of neurotrophins, including brain-derived neurotrophic factor (BDNF) and neurotrophin-4/5 (NT-4/5), in animal models of AD (4). The ability of NGF to prevent the degeneration of basal forebrain cholinergic neurons was also demonstrated in nonhuman primate brain (5) and provided a strong rationale to initiate clinical trials of NGF for AD. The intraventricular infusion of NGF for 3 months in AD subjects resulted in mild cognitive improvements, but adverse effects limited this mode of delivery (6).

A gene therapy strategy for NGF delivery showed greater promise. In a phase I study, autologous fibroblasts genetically modified to express human NGF were grafted into the forebrain. After a mean follow-up of 22 months, there was slight cognitive improvement in 6 subjects and no adverse effects (7). A randomized, controlled phase II trial of NGF gene therapy is ongoing in subjects with mild to moderate AD (ClinicalTrials.gov Identifier: NCT00876863) and results will be eagerly awaited.

Pro- and anti-inflammatory cytokines play an important role in AD, and common polymorphisms of genes controlling their production have been shown to be associated with susceptibility to sporadic AD. For example, functional polymorphisms in the interleukin-18 (IL-18) promoter may be involved in the risk of developing sporadic late-onset AD (8). An interaction associated with the risk of AD has been reported between polymorphisms in the regulatory regions of the genes for the proinflammatory cytokine IL-6 (gene: *IL6*) and the anti-inflammatory cytokine IL-10 (gene: *IL10*) (9). Polymorphisms of the

TNF-α gene (*TNF*) have been reported to affect the risk of developing AD (10). Plasma levels of the hematopoietic cytokine granulocyte colony-stimulating factor (G-CSF) have been found to be significantly reduced in patients with AD, leading researchers to question the role of hematopoietic growth factors (HGFs) in the pathophysiology of the disease (11).

Given the hint of success of neurotrophin therapy in AD, the search for other trophic factors that may impact the degenerating nervous system has expanded to include hematopoietic cytokines or HGFs. Increasingly, researchers have become aware of the overlap between neurotrophins and HGFs. HGFs enhance the proliferation, differentiation and release of all blood cell lineages into the circulation, but surprisingly, HGFs have also been shown to influence the proliferation of neural stem/progenitor cells (NSCs). Review of the similarities and differences between the stem cells that give rise to neural cell lineages ("neuropoiesis") and those that generate the various blood cell lineages (hematopoiesis), led to the concept of the "brain marrow" (12, 13). "Brain marrow" was conceived as a proliferative central core of the central nervous system (CNS) comprising the subependymal zone and other cell groups (subgranular zone of the hippocampal dentate gyrus) that give rise to new neural cells throughout life. In addition to requiring special extracellular matrix and supportive cells (the stem cell "niches"), both sets of stem cells (blood and brain) require trophic factors, cytokines and other molecules that are necessary for proliferation, migration and differentiation into their respective cell lineages. A major overlap between bone and brain marrow is seen in the collection of cytokines and trophic factors that promote blood cell and neural cell development. Many of the cytokines and growth factors required for both hematopoiesis and neuropoiesis are constitutively expressed by bone marrow stromal cells (BMSCs) (12, 14).

Two cytokine superfamilies produced by BMSCs are TGF-α and the hematopoietins. They mediate a range of developmental events in the nervous system that rival those of the classic neurotrophins. The bone morphogenetic proteins (BMPs), a subclass of the TGF-α superfamily, are well known for their significant effects on the development of osteocytes and chondrocytes in bone. However, BMP ligands and their receptor subunits are also present throughout neural development within discrete regions of the embryonic brain and within neural crest-derived migratory zones (15, 16). BMPs exhibit a broad range of cellular and context-specific effects during multiple stages of neural development (17). For example, BMPs initially inhibit the formation of neuroectoderm during gastrulation, while within the neural tube they act as gradient morphogens to promote the differentiation of dorsal and intermediate cell types through cooperative signaling. BMP-2 acts synergistically with fibroblast growth factor (FGF2) on more lineage-restricted embryonic CNS progenitor cells to induce the expression of the dopamine neuronal marker tyrosine hydroxylase (18). Various hematopoietic cytokines have been shown to enhance the number of dopaminergic neurons in mesencephalic cultures, but only IL-1 induced the expression of the dopamine neuronal marker tyrosine hydroxylase in progenitor cells (19).

This review will focus specifically on HGFs that have a direct or indirect effect on the brain and which may have therapeutic potential for neurodegenerative diseases, including AD. A summary of the effects of HGFs on brain that are to be discussed can be found in Table I.

GRANULOCYTE COLONY-STIMULATING FACTOR (G-CSF)

G-CSF is perhaps the most studied and clinically useful of the HGFs. G-CSF is a multimodal cytokine that stimulates hematopoietic stem cell (HSC) proliferation and increases the trafficking of monocytes from the blood to the brain, resulting in increased numbers of activated microglia. G-CSF is anti-inflammatory (20), neuroprotective (21, 22), antiapoptotic (23) and induces the proliferation and differentiation of NSCs into new neurons and astrocytes (24, 25) (Fig. 1). In addition, G-CSF (and other HGFs) promotes cerebral angiogenesis (26, 27), a biological response with implications for AD. Indeed, cerebrovascular dysfunction is associated with and may precede cognitive decline and the onset of neurodegenerative changes in AD and in animal models of AD (28).

G-CSF is a 19-kDa glycoprotein formed into four antiparallel α helices (29). G-CSF stimulates the proliferation and differentiation of HSCs and exists in both 174- and 180-amino-acid forms, although the 174-amino-acid form is more abundant and active and has been used in three proprietary recombinant G-CSF proteins, including lenograstim (Granulocyte®), filgrastim (Neupogen®) and pegylated filgrastim (Neulasta®), which has a longer duration of action (30).

G-CSF has been shown to be synthesized by many cell types, including BMSCs, fibroblasts and macrophages (29, 31, 32) and its receptor G-CSF-R has also been indicated to be just as ubiquitous, as it can be found on the cell surface of endothelial cells, lymphocytes, platelets and neutrophils (33-36). G-CSF and G-CSF-R are also both expressed by neurons in several areas of the brain, including pyramidal cells in cortical layers (specifically II and V), Purkinje cells of the cerebellum, subventricular zone and in cerebellar nuclei in rats. Also, the G-CSF ligand has been identified in cells within the CA3 region of the hippocampus, subgranular zone and the hilus of the dentate gyrus, entorhinal cortex and the olfactory bulb. In postmortem studies G-CSF-R has been found in the human frontal cortex (37).

G-CSF-R is composed of six functional domains which are broken up into an immunoglobulin-like domain, a cytokine receptor-homologous domain (CRH) and three additional fibronectin type III domains within its extracellular region. Various signaling cascades can be initiated by G-CSF binding to G-CSF-R. G-CSF induces the Janus kinase (JAK)/signal transducer and activator of transcription (STAT) pathway, the Ras/mitogen-activated protein (MAP) kinase and the phosphatidylinositol 3-kinase (PI3K)/protein kinase B (PKB/Akt) pathways. These pathways have been shown to induce cellular proliferation, apoptotic processes and anti-inflammatory processes (38-42). Mutations or polymorphisms in the genes encoding for the G-CSF-R and its downstream effectors have been associated with an altered response to G-CSF therapy. Although the clinical implications of such genetic alterations are largely speculative, there is a need for further studies to evaluate whether genetic analysis of the G-CSF-R and/or downstream signaling proteins might tailor the use of G-CSF in patients with neurodegenerative diseases (43).

In a rat stroke model, G-CSF and G-CSF-R were upregulated 2 h after occlusion and reperfusion of the middle cerebral artery (MCA) in the ipsilateral forebrain (37). In another study, G-CSF mRNA levels were greatly elevated in the ischemic brain versus the normal

Table I. *Summary of effects of hematopoietic growth factors (HGFs) in brain.*

SCF (stem cell factor)	G-CSF (granulocyte colony-stimulating factor)	M-CSF (monocyte colony-stimulating factor)	GM-CSF (granulocyte–macrophage colony-stimulating factor)	EPO (erythropoietin)
Survival factor & chemokine of NSCs	Upregulated in ischemic brain	Reduced cognitive deficiency and Aβ plaque load in APP/PS1 Tg mice	Upregulation of its receptor in areas of ischemia in the brain	Its receptor is found in both the nervous & cardiovascular system
Increases neurogenesis	Neurogenesis in AD mice	Decreased Aβ plaque deposition in AD mice	Receptor found on NSCs in hippocampus	Increased levels of EPO and its receptor in the brain with hypoxia or ischemia
Neuroprotective effects	Protective autocrine signaling in response to brain injury	Improvement in spatial memory in AD Tg mice	Antiapoptotic effect on neurons & effect induced by ischemia	Antiapoptotic effects for NSCs against Aβ-induced oxidative stress
Modulation of microglia & possible suppression of microglia to decrease neuroinflammation	Improvement in learning, cognitive function and memory in AD mice	Promotion of proliferation, activation and development of microglia	Decreased infarct size in ischemia & decreased apoptosis in spinal cord with spinal contusion	Prevention of hyperphosphorylation of tau and formation of neurofibrillary tangles
Mobilization of BMSCs into the brain	Reduction of Aβ plaques in hippocampus & entorhinal cortex, as well as decreased soluble Aβ in hippocampus	Low levels in presymptomatic AD in combination with low levels of other HGFs results in progression towards a decrease of cognitive function to dementia	Proliferation & differentiation of microglia	Expression of antiapoptotic genes preventing the activation of caspases and microglia, as well as prevention of phagocytosis of damaged neurons
In brain primarily produced by neurons	Mobilization of bone marrow-derived microglia to Aβ plaque in AD mice	In brain primarily produced by astrocytes	In brain primarily produced by astrocytes or neurons	In brain produced in neurons & astrocytes
Mobilization of neuron-like BMSCs & resident brain NSCs to areas of brain injury such as ischemia	Increased synaptophysin in AD mice		Antibodies to GM-CSF cause decreased Aβ plaque deposits, decreased soluble $A\beta_{1\text{-}42}$ production and microglial activation in AD patients	Increased differentiation of MSCs to neurons and decreased differentiation to glial-like cells in the presence of hypoxia
AChE-I increases SCF in AD patients	Plasma levels decreased in AD patients		Combination of Aβ/GM-CSF/IL-4 antibodies decreased Aβ plaque load	Upregulation of ACh and muscarinic receptors
Increased SCF levels are correlated with increased cognitive function in AD patients	As plasma levels decrease, $A\beta_{1\text{-}42}$ levels increase in the CSF in AD patients		GM-CSF administered in combination with Aβ via adenoviral vectors intranasally in AD mice resulted in lower Aβ levels in the brain	Increased production of glutamine synthase in neuronal-like cells
May increase BDNF				
Modulate spatial memory and hippocampal synaptic potentiation				

NSCs, neural stem cells; BMSCs, bone marrow stem cells; AChE-I, acetylcholinesterase inhibitors; SCF, stem cell factor; AD, Alzheimer's disease; BDNF, brain-derived neurotrophic factor; CSF, cerebrospinal fluid; MSCs, mesenchymal stem cells.

brain and returned to normal after 2 days. This elevation in G-CSF mRNA extended to nonischemic areas of the frontal cortex after photothrombosis in a rat model of focal cerebral ischemia (44). G-CSF in combination with stem cell factor (SCF) administered 10 days after ischemia resulted in improved motor performance, induced transition of bone marrow-derived neuronal cells into the penumbra and proliferation of brain-derived NSCs in neurogenic zones (45). G-CSF may have a protective autocrine signaling mechanism, similar to that seen for other HGFs, such as erythropoietin (EPO), in response to brain injury (46). It is known that G-CSF is able to mobilize $CD34^+$ HSCs and to release them into the blood, allowing them to circulate and migrate to areas of ischemic damage (27, 47-50).

The effects of G-CSF were studied in two mouse models of AD (51). In the acute model aggregated β-amyloid (Aβ) was injected into the hippocampal–cortical junction of the mouse brain, and the chronic model used the Tg2576 AD mouse. Noticeable decreases in learning and memory were seen in the acute model during initial performance in the Morris water maze. After 5 days of dosing of G-CSF,

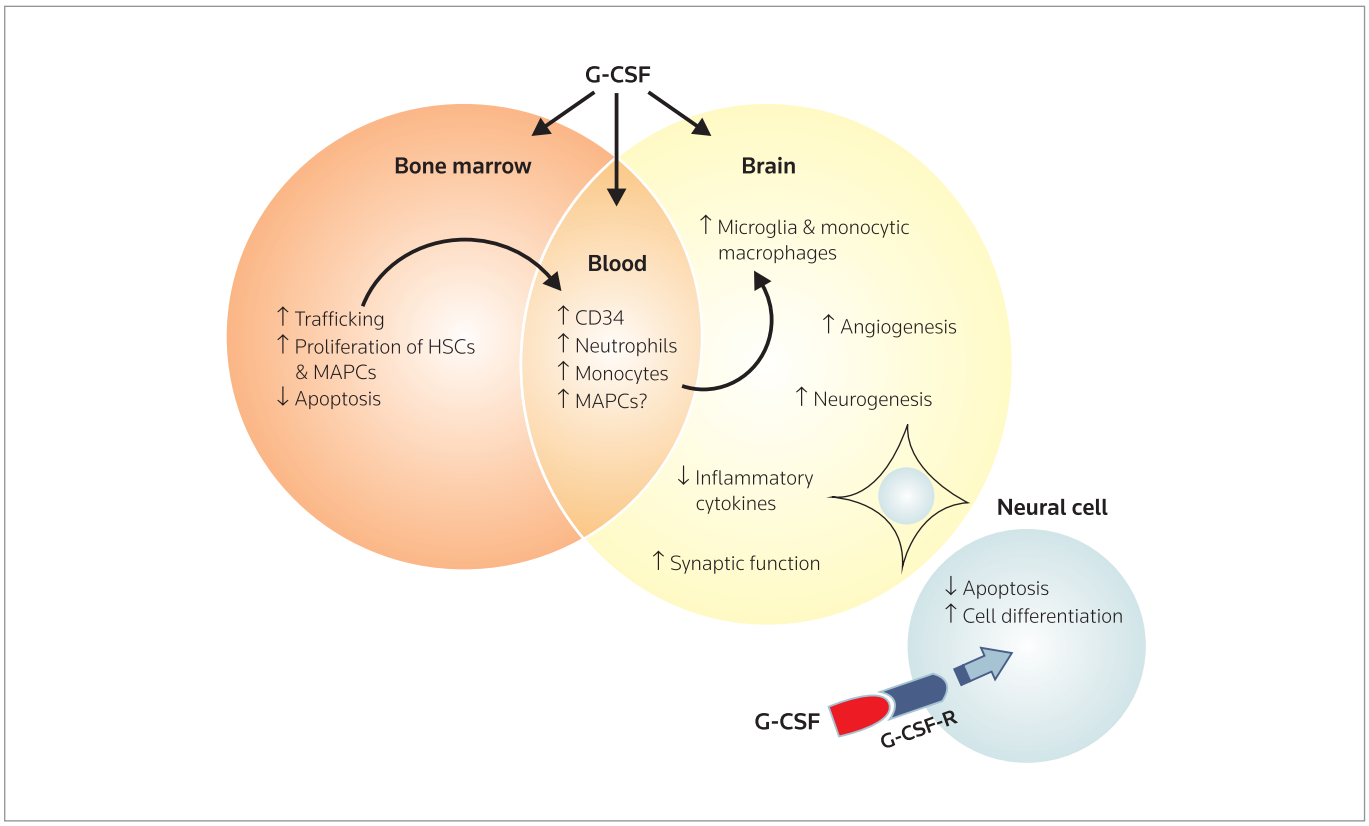

Figure 1. Granulocyte colony-stimulating factor (G-CSF) has direct actions on bone marrow hematopoietic stem cells (HSCs), as well as direct and indirect effects on brain. G-CSF stimulates the proliferation of HSCs and decreases apoptosis in hematopoietic progenitor cells. It also promotes trafficking of CD34 cells and monocytes from marrow to blood. G-CSF promotes the differentiation of the leukocytic lineage to neutrophils. It is also possible that G-CSF stimulates the proliferation of multipotent adult progenitor cells (MAPCs) in the marrow and that this may lead to "transdifferentiation" into neurons, although this process is rare. G-CSF readily accesses the brain parenchyma to activate endogenous microglia. G-CSF also enhances the infiltration of circulating monocytes into the brain, where they differentiate into microglia and monocytic macrophages. G-CSF downregulates inflammatory cytokines in both brain and blood. Within the brain, G-CSF interacts with neural cells bearing G-CSF receptors to inhibit apoptosis, stimulate neurogenesis and synaptogenesis.

learning and memory function was restored to levels similar to those of the control mice. Neurogenesis was increased in both the acute and chronic AD mouse models treated with G-CSF, as seen by the increase in bromodeoxyuridine/neuronal nuclei ($BrdU^+$/NeuN) double-labeling of cells. Twenty percent of the $BrdU^+$ cells in the acute AD mice were $CD34^+$, indicating that a significant proportion of HSCs were mobilized by G-CSF and directed to the Aβ injection site in the brain. Tg2576 mice were examined and found to have significant learning and memory disabilities when performing the Morris water maze and Aβ was found to be present in both the hippocampus and cortex. After 5 days of dosing of G-CSF an improvement in memory and learning was seen by a reduced latency in the Morris water maze, similar to the improvement seen in the acute AD mice treated with G-CSF. Three months after treatment with G-CSF, Tg2576 mice were retested and found to still have significant learning and memory improvement over the untreated mice, indicating a long-lasting effect for G-CSF. Levels of the neurotransmitter ACh were increased by 20% in G-CSF-treated Tg2576 mice. Although learning and memory were improved, no reduction in Aβ plaques was seen. Overall, treatment with G-CSF in both the acute and chronic AD mouse models resulted in release of HSCs from the bone marrow, stimulation of both NSC proliferation and neurogenesis, increased ACh production and an overall improvement in neurological function, specifically improvement in learning and memory.

In a recent study from the author's laboratory, Tg APP/PS1 AD mice were treated with subcutaneous G-CSF injections every other day for 2 weeks (24). A significant reduction of total Aβ deposits in both the hippocampus and entorhinal cortex was seen, as well as a reduction in soluble Aβ within the hippocampus. A correlation was observed between decreased Aβ deposition and improvement in cognitive function. In other studies it has also been shown that an increase in Aβ decreases cognitive function (52, 53). Improved cognitive function was also seen with the removal of Aβ following long-term caffeine administration (54) and administration of leuprolide acetate (a gonadotrophin-releasing hormone [GnRH] analogue) (55). G-CSF also mobilized bone marrow-derived microglia in the Tg mice which targeted Aβ, without causing an additional inflammatory response. It has been noted that in Tg APP/PS1 mice microglia are dysfunctional in their ability to produce the Aβ-binding receptor and Aβ-degrading enzymes (56). Despite the increase in neutrophil count,

G-CSF appears to dampen the release of proinflammatory mediators while anti-inflammatory cytokines are increased (20, 57).

Another interesting observation was the increased synaptophysin immunostaining in the hippocampus, suggesting increased synaptogenesis (24). Increased levels of Aβ have been linked to decreased levels of synapses, but with decreased levels of Aβ the loss of synapses is self-correcting, leading to an improvement in long-term potentiation (LTP), an electrophysiological parameter of learning (58-61). G-CSF was also shown to increase hippocampal neurogenesis in the AD mice, but it is unclear whether or not the neurogenesis was responsible for the improved cognitive performance in the radial arm water maze.

In a recent study, a significant decrease in G-CSF plasma levels was found in patients with early AD in comparison to healthy age-matched controls (11). Also, a significant inverse correlation was shown between G-CSF plasma levels and $A\beta_{1-42}$ levels in the cerebrospinal fluid (CSF) of AD patients. A significant inverse correlation between G-CSF levels and age was seen in both AD patients and healthy controls. Since G-CSF levels decrease with age, it would be interesting to assess if G-CSF levels decrease as the severity of AD progresses.

G-CSF is currently being used therapeutically to induce the generation of white blood cells (WBCs) in patients suffering from neutropenia after anticancer therapy, as well as in patients with congenital neutropenia (62-64). Given that G-CSF is already in clinical use, it has been suggested that it could be used to treat various neurological disorders without compromising the safety of the patient. Clinical trials to examine the ability of G-CSF to treat ischemic strokes are currently under way (65). Additionally, G-CSF has been used to treat myocardial infarction (66-70). Very recently, a clinical trial to study the efficacy and safety of G-CSF as a procognitive agent was initiated in mild to moderate AD at the University of South Florida (71). This is a tolerability and safety study with a secondary objective of detecting a procognitive benefit.

G-CSF has a long track record of safety. A recent prospective study of the safety of G-CSF in 1,278 normal donors found that 870 donors (68%) presented with some G-CSF-related toxicity. Bone pain was observed most frequently (784 donors, 90%), followed by headache (290 donors, 33%), fever (56 cases, 6%), fatigue (6%) and nausea (5%) (72). Bone pain was effectively treated with common analgesics such as acetaminophen or ibuprofen, and the symptoms generally resolved within 1 week after discontinuation of G-CSF administration. Other less common effects attributed to G-CSF were sweating and insomnia, which were observed in 19 (2.2%) and 16 (1.3%) donors, respectively. Two donors with previous hyperuricemia had gout crises during the mobilization procedure. There were no mobilization-related deaths, although some rare but serious side effects of G-CSF were reported. One donor had a splenic rupture 12 h after the last leukapheresis, which required splenectomy.

Several other studies have evaluated the effects of short–term administration of G-CSF on the spleen in normal donors and patients with cancer or neutropenia. These studies showed a median increase of 11 mm in spleen length in G-CSF recipients. In the majority of the cases, the spleen length increase was transient and reversible (73). At least four cases of splenic rupture have been associated with an increase in spleen size in healthy adult donors. A retrospective study on G-CSF-associated adverse events in healthy individuals or subjects with chronic neutropenia or cancer did not find an increased risk of malignancy among healthy individuals who received G-CSF before peripheral blood stem cell harvesting. However, more than 2,000 donors would have to be followed for 10 years to detect a 10-fold increase in leukemia risk (74).

STEM CELL FACTOR (SCF)

SCF is an 18.5-kDa glycoprotein that plays a key role in hematopoiesis. It is also known as c-kit ligand, steel factor or mast cell growth factor. Its structure is similar to monocyte colony-stimulating factor (M-CSF). SCF exists in both soluble (sSCF) and membrane-bound (mSCF) forms. sSCF activates the c-kit (SCF) receptor more transiently and downregulation is induced more rapidly than in mSCF. mSCF may participate in guiding the HSCs to their final destinations in the body during embryogenesis and be biologically more active than sSCF (75). SCF and its receptor are found in organs throughout the body, but of particular interest, both are expressed in the neuroproliferative zones of the brain (76-78). Mutations or genetic polymorphisms resulting in alteration of c-kit function may alter the therapeutic effects of SCF and also result in specific diseases (79).

Several biological functions have been observed for SCF in vitro and in vivo. In vitro, SCF has been shown to increase the survival of HSCs, generate direct colony-forming cells, potentiate the ability of epinephrine and ADP to stimulate platelet aggregation, assist in serotonin (5-HT) secretion, promote melanogenesis and gametogenesis, have synergistic effects on megakaryocytes and support the growth and function of mast cells. In vivo, SCF has been shown to participate in SC self-renewal, have a synergistic effect with other HGFs such as G-CSF, have chemotactic effects for SCs, assist in the development of early T cells and mobilize HSCs from the bone marrow to the blood (75). Its specific functions in the CNS include acting as a survival factor and chemokine for NSCs (80), neurogenesis (81), neuroprotective effects (82) and modulation of microglia (83). In the normal brain, SCF is produced primarily by neurons and not glia (84), and systemic administration of SCF enhances infiltration of BMSCs into the brain (85). In the injured brain, SCF expression is upregulated (84) and administration of SCF to ischemic brain mobilizes neuron-like bone marrow-derived cells and resident brain NSCs to areas of injury, especially the penumbra (45, 84, 86).

Interestingly, treatment of AD patients with donepezil (AChE-I) may not only act by increasing cholinergic synaptic activity through an increase of ACh, but also through an increase in HGFs, such as BDNF and SCF (87). AD patients with higher levels of SCF had a higher level of cognitive function or a smaller loss of cognitive function than those with lower levels of SCF. AChE-I have been shown to activate protein kinase C (PKC), which cleaves mSCF, releasing it from the cell surface. This increase in SCF may be responsible for the increase of BDNF and the decrease in neuroinflammation through the suppression of microglia (88).

In c-kit mutant rats a decrease in escape latency with repeated trials of the water maze was not shown, in contrast to wild-type mice, and they displayed impairment in spatial memory. The mutant rats also

showed impairment in hippocampal synaptic potentiation in comparison to wild-type rats in studies of paired-pulse facilitation (PPF) in the mossy fiber-CA3 hippocampal pathway and LTP (89). Therefore, a lack of SCF impairs both spatial memory and hippocampal synaptic potentiation or long-term memory.

Lower levels of SCF have been observed in plasma of AD patients in comparison to age-matched healthy controls. Also, lower levels of plasma SCF have been significantly correlated with an increase in the severity of dementia. Patients with AD had significantly lower CSF levels of SCF compared to those with noninflammatory neurological disease (NIND). No correlation exists between the levels of SCF found in the CSF and plasma, so the SCF in these locations may have different sources (90). A decrease in HGFs, such as SCF, could contribute to a decrease in HSCs in patients with AD, although due to the negative feedback loop between sSCF and mSCF it is possible that sSCF is decreasing while mSCF is increasing (91).

Both SCF and FGF2 protein and mRNA levels are increased with hypoxia in cell culture. Also, SCF is increased in immature neurons in both the subgranular zone and the subventricular zone in rat brain, and administration of SCF increases BrdU labeling of immature neurons, showing the effect of SCF in neurogenic zones (81). In those cells adjacent to the areas of brain injury, higher levels of SCF are expressed than those that are more distant from the site of injury, and SCF mRNA is overexpressed in the injured area of the brain (84).

In subacute brain ischemia the administration of SCF + G-CSF decreased infarct size and resulted in functional improvement (45). SCF + G-CSF treatment in a mouse model of chronic stroke increased BMSC mobilization and differentiation, resulting in an increase in neurogenesis and angiogenesis, and therefore enhancement of brain repair in ischemia (92). Both SCF and G-CSF pass through the intact blood–brain barrier (BBB) (86). Cerebral hypoperfusion and impaired Aβ clearance across the BBB may contribute to the onset and progression of AD. Accumulation of Aβ in cerebral blood vessels is associated with cognitive decline and can result in disruption of the blood vessel wall, leading to micro- or macrointracerebral bleeds that exacerbate the neurodegenerative process and inflammatory response (28). Given the ability of SCF and G-CSF to increase angiogenesis and ameliorate the neurovascular system in animal models of stroke, it is likely that this mechanism may be involved in mediating the beneficial effects of HGFs in AD.

Although SCF may be a potential treatment for neurological diseases, such as AD and stroke, it has also been shown to have side effects in early clinical trials. It can cause wheals at the site of injection due to activation of mast cells and darkening in pigmentation due to activation of melanocytes, although if given at lower doses in conjunction with a synergistic factor such as G-CSF these side effects may be eliminated (75). Recently, phase I clinical trials have been completed with SCF as a treatment for aplastic anemia, myelodysplasia and AIDS/AIDS-related malignancies.

GRANULOCYTE–MACROPHAGE COLONY-STIMULATING FACTOR (GM-CSF)

GM-CSF is a 23-kDa glycoprotein with a four-helical-bundle structure (93) and promotes the maturation and differentiation of HSCs into granulocytes and macrophages. GM-CSF stimulates the progression of the cell cycle by upregulating cyclins, and results in proliferation of microglia and their transformation into the ameboid (active) form (94). In the brain, GM-CSF is expressed by astrocytes (32), neurons and, to a lesser extent, oligodendrocytes (95).

The GM-CSF receptor (GM-CSF-R) is heterodimeric and contains an α-chain, the GM-CSF ligand binder, and a β-chain, the signal transducer which is coupled to the STAT pathway (93, 96). GM-CSF-R is expressed by hematopoietic cells, as well as neurons (97). In the uninjured brain, GM-CSF-R is found throughout the brain in areas such as the hippocampal CA2 and CA3 layers, entorhinal cortex and pyramidal cells in the cortex, as well as several other areas. GM-CSF-R can also be detected in some oligodendrocytes, but not microglia or astrocytes (95), although other researchers claim that GM-CSF-R is also expressed by astrocytes (98). GM-CSF-R and nestin, a marker of NSCs, are coexpressed in the subgranular zone of the hippocampus (99). Following experimental cerebral ischemia, there is an upregulation of the GM-CSF-R in large pyramidal neurons located in the penumbra, indicating an adaptive response to neuronal damage (95).

In the peripheral nervous system, GM-CSF stimulates phagocytosis of myelin during Wallerian degeneration (WD), but this is inhibited at a later stage of WD by a binding protein (100, 101). In the CNS GM-CSF causes the proliferation, induction and differentiation of microglia (102, 103) and has a strong antiapoptotic effect on recruited neutrophils and neurons (95, 104). GM-CSF induces this antiapoptotic effect through an increase in the antiapoptotic proteins Bcl-XL and Bcl-2, and partially through activation of the Akt pathway; this antiapoptotic effect can be activated by stimuli such as an ischemic insult. In two rat models of stroke GM-CSF was administered i.v. and a decrease in infarct size was seen in both models (95). GM-CSF treatment in the disease model of spinal contusion resulted in decreased apoptosis within the spinal cord (105). Treatment of NSCs in vitro with GM-CSF increased differentiation of the stem cells into neurons, evidenced by a significant increase in the numbers of cells expressing the neuronal markers tubulin beta-III and microtubule-associated protein 2 (MAP-2) (99).

Treatment of Tg2576 AD mice with an anti-GM-CSF antibody resulted in a significant decrease in Aβ plaque deposits, soluble $A\beta_{1\text{-}42}$ production, and a decrease in microglial activation (106). Since decreased levels of cytokines such as GM-CSF are associated with a decrease in microglia, the increased GM-CSF levels in AD may contribute to microgliosis (107). Administration of GM-CSF and Aβ via adenoviral vectors intranasally to Tg2576 mice resulted in lower Aβ levels in the brain compared to those not receiving the treatment (108). Tg amyloid precursor protein (APP) mice treated with a combination of Aβ/GM-CSF/IL-4 antibodies showed a significant decrease in the Aβ plaque load (109).

In patients with stroke, AD and vascular dementia an increase in GM-CSF levels has been observed in the CSF and plasma, except for stroke which does not show an elevation in plasma. The increase in GM-CSF may be due to increased neuronal damage and cell death, as a positive correlation was shown between markers for neuronal damage such as *TAU* and *FAS* genes (107, 110, 111). GM-CSF has recently been identified as a neuronal growth factor in the rat brain that is both antiapoptotic and reduces infarct size in vivo (95).

Clinically, GM-CSF has been used for the treatment of neutropenia. GM-CSF, as well as G-CSF, is in clinical trials for the treatment of stroke and appears to be safe. Given their availability, relative safety and beneficial impact on the neurovascular system, some investigators advocate fast-tracking these agents for the treatment of neurodegenerative diseases such as AD.

MONOCYTE COLONY-STIMULATING FACTOR (M-CSF)

M-CSF (also known as CSF-1) is another HGF that is an 80-100-kDa glycoprotein or a 130-160-kDa chondroitin sulfate-containing proteoglycan, or expressed as a membrane-bound 68-86-kDa glycoprotein on M-CSF-producing cells. In all its active forms it is dimeric (112). M-CSF is produced by astrocytes (113). M-CSF has been shown to reduce cognitive deficits and Aβ plaque load in APP/PS1 (presenilin 1) Tg mice, as demonstrated by decreased escape latencies in repeated T-water maze trials in Tg mice receiving i.p. injections of M-CSF and wild-type mice versus Tg mice receiving saline showing an improvement in spatial memory with M-CSF. Tg mice receiving M-CSF injections at 6 months of age had an improved quality of nest building, reflecting diminished social withdrawal and apathy as compared to saline-treated Tg mice. Also, M-CSF-treated Tg mice had a higher ratio of microglia per plaque than control Tg animals. The increase in microglia per plaque may be related to the decreased density, size and overall Aβ burden in the plaques seen in the M-CSF-treated Tg mice (114).

It has also been shown that M-CSF can promote the proliferation, activation and development of microglia, and mice with decreased levels of M-CSF therefore exhibit lower numbers of microglia (113, 115, 116). M-CSF induces proliferation of microglia in culture without affecting the microglial response to lipopolysaccharide (LPS) or Aβ; hence, M-CSF can be used to improve microglial yield in culture without introducing activation artefacts (117). Low levels of M-CSF in presymptomatic AD in combination with low levels of other HGFs are associated with a progression towards a decrease in cognitive function to dementia (118).

STROMAL CELL-DERIVED FACTOR 1 (SDF-1)

SDF-1, also known as C-X-C motif chemokine 12 (CXCL12), is a member of the C-X-C subfamily of chemokines and thus a potent chemoattractant for HSCs that is expressed in the bone marrow (119-122). SDF-1 and its receptor CXCR4 may play a role in blood vessel growth and development, as well as attraction of HSCs to injured vessel walls (123-125). SDF-1 plays an important role in the mobilization, differentiation and adhesion of HSCs and has neuroprotective effects in the CNS (125-127). Decreased plasma levels of SDF-1 have been seen in patients with AD (128) and low levels of SDF-1 may therefore further contribute to the deficient profile of HSC support in the AD brain.

ERYTHROPOIETIN (EPO)

EPO is a 30-kDa hematopoietic cytokine consisting of 165 amino acids and coded by a single copy on chromosome 7 (129, 130). The biological activity of EPO appears to be dependent on its glycosylated chains, as well as the disulfide bonds between the cysteine residues located at positions 7 and 160 and 29 and 33 (131). EPO is ubiquitous throughout the body and is primarily produced in the kidney by peritubular fibroblasts in the renal cortex (132, 133). The main actions of EPO include promotion of red blood cell (RBC) survival through the prevention of apoptosis in threatened RBCs and stimulation of colony-forming erythroid cells to proliferate into new RBCs (134, 135). The production of EPO also occurs in the liver by Kupffer cells and hepatocytes, as well as in the uterus (136). Surprisingly, EPO production and the expression of the erythropoietin receptor (EPO-R) are found in cells of the nervous and cardiovascular systems, suggesting that EPO may have neurovascular protective benefits (129).

The expression of EPO and its receptor have been shown to correlate with decreased levels of oxygen or increased metabolic stress, and to be under the control of hypoxia-inducible factor 1 (HIF-1). HIF-1 is constitutively produced in cells, but undergoes elimination through ubiquitination and proteasome degradation in the presence of oxygen. In hypoxic events, however, the degradation of HIF-1 is inhibited, allowing it to upregulate the expression of both EPO and EPO-R. The timely appearance of EPO and EPO-R in the event of a decrease in oxygen allows EPO to protect cells from apoptosis (129, 130). EPO is currently used to treat anemia in various clinical scenarios (137).

Neurons and astrocytes synthesize EPO and also express the EPO-R on their cellular membranes (138-141). EPO is able to cross the BBB through receptor-mediated transport, allowing EPO produced in peripheral tissues to enter the brain (138). This event, however, only accounts for 6-8% of the total EPO concentration found in the CSF, indicating that most of the EPO in the CSF is produced in the brain (129). Various regions of the brain are considered to be involved in the production of EPO, such as the hippocampus, cortex and midbrain (129, 130). The presence of EPO and EPO-R is relatively low in normal brain and only after metabolic stress or hypoxia do their levels increase, as demonstrated by increases in EPO levels in the CSF, as well as in postmortem brains that have sustained injury, hemorrhage or stroke (142-145).

EPO-R expression is increased in chronic cerebrovascular diseases, such as AD, suggesting that the AD brain is under hypoxic or metabolic stress (146). Aβ plaques are believed to increase oxidative stress and Aβ-induced apoptosis associated with AD (147-150). EPO is able to attenuate the apoptosis induced by reactive oxygen species (ROS) induced by Aβ. Aβ was shown to decrease the ratio of Bcl-2/BAX, leading to activation of caspase-3 and eventually apoptosis. However, EPO was able to reverse this ratio by increasing the expression of the antiapoptotic gene *BCL2* and by decreasing the expression of the proapoptotic gene *BAX*, thus leading to protection of PC-12 cells from Aβ-induced oxidative stress. This was linked to the activation of JAK2/PI3K/Akt pathways, as indicated by an increase in phosphorylated Akt following treatment of PC-12 cells with EPO (151, 152).

The fragmentation of DNA through caspases is a relatively late-occurring phenomenon, as many earlier stimuli also lead to phagocytosis or apoptosis of cells in the CNS. Aβ plaques are able to induce microglial secretion of TNF-α, as well as other inflammatory cytokines (153, 154). Aβ plaques also cause the early expression of externalized phosphatidylserine (PS), signaling microglia to phagocytize the compromised neurons (155-157). EPO has been shown to provide long-term survival through Akt activation, causing the

translocation and activation of nuclear factor NF-kappa-B (NF-κB) to the nucleus, leading to expression of antiapoptotic genes and thus preventing TNF-α-induced apoptosis and the activation of caspase pathways, as well as preventing the externalization of PS that would normally lead to microglial activation and phagocytosis of damaged neurons (158-160).

EPO may also have an impact on the phosphorylation status of tau, a component of neurofibrillary tangles. Hyperphosphorylation of tau protein by glycogen synthase kinase-3 β (GSK-3β) causes destabilization of the cytoskeleton, resulting in the cell death seen in AD (161, 162). GSK-3β phosphorylation of tau is carried out through a reduction in Akt activity, leading to activation of GSK-3β. On the other hand, EPO was shown to induce dephosphorylation and deactivation of GSK-3β by EPO binding its receptor and upregulating Akt activity through the JAK2/PI3K pathway, thus preventing hyperphosphorylation of tau protein and subsequent tangle formation. This provided a possible avenue for preventing Aβ-induced cellular death in the brain (152, 163).

In addition to its neuroprotective capacity, EPO provides the necessary signaling to cause mesenchymal stem cells (MSCs) to differentiate into neurons during simultaneous hypoxia (164, 165). In the presence of EPO, MSCs have been shown to increase the expression of neural markers, such as choline acetyltransferase (ChAT), NeuN and muscarinic receptors, and to increase functional markers, such as synaptophysin, the glutamate/aspartate transporter (GLAST) and neprilysin (164). EPO has also been shown to upregulate the production of ACh and, along with the increase in muscarinic receptors, it appears that these MSCs can be used to replace ACh neurons, which are often associated with cognitive loss in AD (164, 166). It has also been observed that EPO not only increases the differentiation of MSCs into neurons, but also inhibits their differentiation into glial-like cells, evidenced by a decrease in the number of glial fibrillary acidic protein (GFAP)-positive cells following EPO administration. EPO-treated MSCs are able to produce glutamine synthase (GS), which is normally produced by astrocytes to combat glutamate toxicity, but GS activity is decreased in AD, which causes neurons to synthesize it (164, 167). Along with EPO induction of GLAST and GLT-1 production, it has been suggested that EPO-treated MSCs can take over the task of metabolizing glutamate that is normally performed by astrocytes (164, 168). It is therefore hypothesized that coadministration of transplanted MSCs and EPO could offer a possible therapeutic option for the treatment of AD (164).

CONCLUSIONS

The application of HGFs for the treatment of AD is based on a growing body of strong preclinical data and the fact that many of these agents are already in clinical use in the field of hematology/oncology. In the past 10 years, it has become apparent that hematopoiesis and neuropoiesis have much in common, especially the trophic factor/cytokines that regulate self-renewal and differentiation into blood cell lineages and neural cell lineages. Bone marrow houses more "primitive", multipotent adult progenitor cells (MAPCs) that are capable of giving rise to tissues of all embryonic germ layers. Bone marrow-derived cells have been shown by independent investigators to give rise to neural cells in vitro, and in vivo studies have demonstrated that bone marrow cells migrate into the brain, where they appear to differentiate into neurons and glia. The mechanism for transdifferentiation of bone marrow cells to neural cells is not clear and may reflect the capacity of bone marrow-derived cells to fuse with injured neurons. In vivo studies suggest that the generation of new neurons from BMSC/progenitor cells, if it really occurs, may be a rare phenomenon. Nevertheless, bone marrow-derived cells from the peripheral circulation are the source of at least 10% of the adult brain's immune cells (microglia) under normal conditions, and following various brain injuries, the trafficking of bone marrow cells from blood to brain is markedly increased. Microglia have been clearly observed in the core of amyloid plaques in transgenic mouse models of AD. Perhaps as many as 10% of these cells originate from the bone marrow. The $A\beta_{1\text{-}40}$ and $A\beta_{1\text{-}42}$ isoforms are among the many signals that trigger this chemoattraction. HGFs also modulate the humoral neuroimmune response: G-CSF decreases proinflammatory cytokines and upregulates anti-inflammatory cytokines. Moreover, G-CSF and other HGFs have direct actions on the CNS. They inhibit programmed cell death and promote neurogenesis and synaptogenesis. Administration of G-CSF has been shown to be safe and well tolerated in early clinical studies in patients with ischemic stroke. There is likely to be a surge in the development of clinical trials of G-CSF, as well as other HGFs, to reverse dementia or forestall the progression of AD.

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DISCLOSURE

The authors state no conflicts of interest.

REFERENCES

1. DeKosky, S.T., Scheff, S.W., Styren, S.D. *Structural correlates of cognition in dementia: Quantification and assessment of synapse change.* Neurodegeneration 1996, 5(4): 417-21.
2. Selkoe, D.J. *Alzheimer's disease is a synaptic failure.* Science 2002, 298(5594): 789-91.
3. Swaab, D.F., Dubelaar, E.J., Hofman, M.A., Scherder, E.J., van Someren, E.J., Verwer, R.W. *Brain aging and Alzheimer's disease; use it or lose it.* Prog Brain Res 2002, 138: 343-73.
4. Schulte-Herbruggen, O., Jockers-Scherubl, M.C., Hellweg, R. *Neurotrophins: From pathophysiology to treatment in Alzheimer's disease.* Curr Alzheimer Res 2008, 5(1): 38-44.
5. Tuszynski, M.H., Sang, H., Yoshida, K., Gage, F.H. *Recombinant human nerve growth factor infusions prevent cholinergic neuronal degeneration in the adult primate brain.* Ann Neurol 1991, 30(5): 625-36.
6. Eriksdotter Jonhagen, M., Nordberg, A., Amberla, K. et al. *Intracerebroventricular infusion of nerve growth factor in three patients with Alzheimer's disease.* Dement Geriatr Cogn Disord 1998, 9(5): 246-57.
7. Tuszynski, M.H., Thal, L., Pay, M. et al. *A phase 1 clinical trial of nerve growth factor gene therapy for Alzheimer disease.* Nat Med 2005, 11(5): 551-5.
8. Yu, J.T., Tan, L., Song, J.H., Sun, Y.P., Chen, W., Miao, D., Tian, Y. *Interleukin-18 promoter polymorphisms and risk of late onset Alzheimer's disease.* Brain Res 2009, 1253: 169-75.

9. Combarros, O., Van Duijin, C.M., Hammond, N. et al. *Replication by the epistasis project of the interaction between the genes for IL-6 and IL-10 in the risk of Alzheimer's disease.* J Neuroinflammation 2009, 6: 22.
10. Di Bona, D., Candore, G., Franceschi, C. et al. *Systematic review by meta-analyses on the possible role of TNF-alpha polymorphisms in association with Alzheimer's disease.* Brain Res Rev 2009, 61(2): 60-8.
11. Laske, C., Stellos, K., Stransky, E., Leyhe, T., Gawaz, M. *Decreased plasma levels of granulocyte-colony stimulating factor (G-CSF) in patients with early Alzheimer's disease.* J Alzheimers Dis 2009, 17(1): 115-23.
12. Scheffler, B., Horn, M., Blumcke, I., Laywell, E.D., Coomes, D., Kukekov, V.G., Steindler, D.A. *Marrow-mindedness: A perspective on neuropoiesis.* Trends Neurosci 1999, 22: 348-57.
13. Song, S., Sanchez-Ramos, J. *Brain as the sea of marrow.* Exp Neurol 2003, 184(1): 54-60.
14. Sanchez-Ramos, J. et al. *The potential of bone marrow as a source of neural precursors.* Neurosci News 2000, 3: 32-43.
15. Gamer, L.W., Wolfman, N.M., Celeste, A.J., Hattersley, G., Hewick, R., Rosen, V. *A novel BMP expressed in developing mouse limb, spinal cord, and tail bud is a potent mesoderm inducer in Xenopus embryos.* Dev Biol 1999, 208(1): 222-32.
16. Mehler, M.F., Kessler, J.A. *Growth factor regulation of neuronal development.* Dev Neurosci 1994, 16(3-4): 180-95.
17. Mehler, M.F., Mabie, P.C., Zhang, D., Kessler, J.A. *Bone morphogenetic proteins in the nervous system.* Trends Neurosci 1997, 20(7): 309-17.
18. Daadi, M., Arcellana-Panlilio, M.Y., Weiss, S. *Activin co-operates with fibroblast growth factor 2 to regulate tyrosine hydroxylase expression in the basal forebrain ventricular zone progenitors.* Neuroscience 1998, 86(3): 867-80.
19. Ling, Z.D., Potter, E.D., Lipton, J.W., Carvey, P.M. *Differentiation of mesencephalic progenitor cells into dopaminergic neurons by cytokines.* Exp Neurol 1998, 149(2): 411-23.
20. Hartung, T. *Anti-inflammatory effects of granulocyte colony-stimulating factor.* Curr Opin Hematol 1998, 5(3): 221-5.
21. Lu, C.Z., Xiao, B.G. *Neuroprotection of G-CSF in cerebral ischemia.* Front Biosci 2007, 12: 2869-75.
22. Jung, K.H., Chu, K., Lee, S.T., Kang, L., Kim, S.U., Kim, M., Roh, J.K. *G-CSF protects human cerebral hybrid neurons against in vitro ischemia.* Neurosci Lett 2006, 394(3): 168-73.
23. Yata, K., Matchett, G.A., Tsubokawa, T., Tang, J., Kanamaru, K., Zhang, J.H. *Granulocyte-colony stimulating factor inhibits apoptotic neuron loss after neonatal hypoxia-ischemia in rats.* Brain Res 2007, 1145: 227-38.
24. Sanchez-Ramos, J., Song, S., Sava, V. et al. *Granulocyte colony stimulating factor decreases brain amyloid burden and reverses cognitive impairment in Alzheimer's mice.* Neuroscience 2009, 163(1): 55-72.
25. Jung, K.H., Chu, K., Lee, S.T. et al. *Granulocyte colony-stimulating factor stimulates neurogenesis via vascular endothelial growth factor with STAT activation.* Brain Res 2006, 1073-1074: 190-201.
26. Toth, Z.E., Leker, R.R., Shahar, T. et al. *The combination of granulocyte colony-stimulating factor and stem cell factor significantly increases the number of bone marrow-derived endothelial cells in brains of mice following cerebral ischemia.* Blood 2008, 111(12): 5544-52.
27. Shyu, W.C., Lin, S.Z., Yang, H.I., Tzeng, Y.S., Pang, C.Y., Yen, P.S., Li, H. *Functional recovery of stroke rats induced by granulocyte colony-stimulating factor-stimulated stem cells.* Circulation 2004, 110(13): 1847-54.
28. Bell, R.D., Zlokovic, B.V. *Neurovascular mechanisms and blood-brain barrier disorder in Alzheimer's disease.* Acta Neuropathol 2009, 118(1): 103-13.
29. Wells, J.A., de Vos, A.M. *Hematopoietic receptor complexes.* Annu Rev Biochem 1996, 65: 609-34.
30. Sanchez-Ramos, J., Song, S., Cao, C., Arendash, G. *The potential of hematopoietic growth factors for treatment of Alzheimer's disease: A mini-review.* BMC Neurosci 2008, 9(Suppl. 2): S3.
31. Demetri, G.D., Griffin, J.D. *Granulocyte colony-stimulating factor and its receptor.* Blood 1991, 78(11): 2791-808.
32. Malipiero, U.V., Frei, K., Fontana, A. *Production of hemopoietic colony-stimulating factors by astrocytes.* J Immunol 1990, 144(10): 3816-21.
33. Bussolino, F., Wang, J.M., Defilippi, P. et al. *Granulocyte- and granulocyte-macrophage-colony stimulating factors induce human endothelial cells to migrate and proliferate.* Nature 1989**,** 337(6206): 471-3.
34. Morikawa, K., Morikawa, S., Nakamura, M., Miyawaki, T. *Characterization of granulocyte colony-stimulating factor receptor expressed on human lymphocytes.* Br J Haematol 2002, 118(1): 296-304.
35. Shimoda, K., Okamura, S., Harada, N., Kondo, S., Okamura, T., Niho, Y. *Identification of a functional receptor for granulocyte colony-stimulating factor on platelets.* J Clin Invest 1993, 91(4): 1310-3.
36. Hanazono, Y., Hosoi, T., Kuwaki, T., Matsuki, S., Miyazono, K., Miyagawa, K., Takaku, F. *Structural analysis of the receptors for granulocyte colony-stimulating factor on neutrophils.* Exp Hematol 1990, 18(10): 1097-103.
37. Schneider, A., Krüger, C., Steigleder, T. et al. *The hematopoietic factor G-CSF is a neuronal ligand that counteracts programmed cell death and drives neurogenesis.* J Clin Invest 2005, 115(8): 2083-98.
38. Hunter, M.G., Avalos, B.R. *Phosphatidylinositol 3'-kinase and SH2-containing inositol phosphatase (SHIP) are recruited by distinct positive and negative growth-regulatory domains in the granulocyte colony-stimulating factor receptor.* J Immunol 1998, 160(10): 4979-87.
39. Shimoda, K., Feng, J., Murakami, H. et al. *Jak1 plays an essential role for receptor phosphorylation and Stat activation in response to granulocyte colony-stimulating factor.* Blood 1997, 90(2): 597-604.
40. Tian, S.S., Lamb, P., Seidel, H.M., Stein, R.B., Rosen, J. *Rapid activation of the STAT3 transcription factor by granulocyte colony-stimulating factor.* Blood 1994, 84(6): 1760-4.
41. Ward, A.C., Loeb, D.M., Soede-Bobok, A.A., Touw, I.P., Friedman, A.D. *Regulation of granulopoiesis by transcription factors and cytokine signals.* Leukemia 2000, 14(6): 973-90.
42. Dong, F., Larner, A.C. *Activation of Akt kinase by granulocyte colony-stimulating factor (G-CSF): Evidence for the role of a tyrosine kinase activity distinct from the Janus kinases.* Blood 2000, 95(5): 1656-62.
43. Crea, F., Giovannetti, E., Zinzani, P.L., Danesi, R. *Pharmacologic rationale for early G-CSF prophylaxis in cancer patients and role of pharmacogenetics in treatment optimization.* Crit Rev Oncol Hematol 2009, 72(1): 21-44.
44. Kleinschnitz, C., Schroeter, M., Jander, S., Stoll, G. *Induction of granulocyte colony-stimulating factor mRNA by focal cerebral ischemia and cortical spreading depression.* Mol Brain Res 2004, 131(1-2): 73-8.
45. Kawada, H., Takizawa, S., Takanashi, T. et al. *Administration of hematopoietic cytokines in the subacute phase after cerebral infarction is effective for functional recovery facilitating proliferation of intrinsic neural stem/progenitor cells and transition of bone marrow-derived neuronal cells.* Circulation 2006, 113(5): 701-10.
46. Solaroglu, I., Jadhav, V., Zhang, J.H. *Neuroprotective effect of granulocyte-colony stimulating factor.* Front Biosci 2007, 12: 712-24.
47. Bodine, D.M., Seidel, N.E., Gale, M.S., Nienhuis, A.W., Orlic, D. *Efficient retrovirus transduction of mouse pluripotent hematopoietic stem cells mobilized into the peripheral blood by treatment with granulocyte colony-stimulating factor and stem cell factor.* Blood 1994, 84(5): 1482-91.

48. Orlic, D., Kajstura, J., Chimenti, S. et al. *Mobilized bone marrow cells repair the infarcted heart, improving function and survival.* Proc Natl Acad Sci U S A 2001, 98(18): 10344-9.
49. Orlic, D., Kajstura, J., Chimenti, S. et al. *Bone marrow cells regenerate infarcted myocardium.* Nature 2001, 410(6829): 701-5.
50. Grigg, A.P., Roberts, A.W., Raunow, H. et al. *Optimizing dose and scheduling of filgrastim (granulocyte colony-stimulating factor) for mobilization and collection of peripheral blood progenitor cells in normal volunteers.* Blood 1995, 86(12): 4437-45.
51. Tsai, K.J., Tsai, Y.C., Shen, C.K. *G-CSF rescues the memory impairment of animal models of Alzheimer's disease.* J Exp Med 2007, 204(6): 1273-80.
52. Arendash, G.W., King, D.L., Gordon, M.N., Morgan, D., Hatcher, J.M., Hope, C.E., Diamond, D.M. *Progressive, age-related behavioral impairments in transgenic mice carrying both mutant amyloid precursor protein and presenilin-1 transgenes.* Brain Res 2001, 891(1-2): 42-53.
53. Ethell, D.W., Shippy, D., Cao, C., Cracchiolo, J.R., Runfeldt, M., Blake, B., Arendash, G.W. *Abeta-specific T-cells reverse cognitive decline and synaptic loss in Alzheimer's mice.* Neurobiol Dis 2006, 23(2): 351-61.
54. Arendash, G.W., Schleif, W., Rezai-Zadeh, K. et al. *Caffeine protects Alzheimer's mice against cognitive impairment and reduces brain beta-amyloid production.* Neuroscience 2006, 142(4): 941-52.
55. Casadesus, G., Webber, K.M., Atwood, C.S., Pappolla, M.A., Perry, G., Bowen, R.L., Smith, M.A. *Luteinizing hormone modulates cognition and amyloid-beta deposition in Alzheimer APP transgenic mice.* Biochim Biophys Acta 2006, 1762(4): 447-52.
56. Hickman, S.E., Allison, E.K., El Khoury, J. *Microglial dysfunction and defective beta-amyloid clearance pathways in aging Alzheimer's disease mice.* J Neurosci 2008, 28(33): 8354-60.
57. Boneberg, E.M., Hareng, L., Gantner, F., Wendel, A., Hartung, T. *Human monocytes express functional receptors for granulocyte colony-stimulating factor that mediate suppression of monokines and interferon-gamma.* Blood 2000, 95(1): 270-6.
58. Hsia, A.Y., Masliah, E., McConlogue, L. et al. *Plaque-independent disruption of neural circuits in Alzheimer's disease mouse models.* Proc Natl Acad Sci U S A 1999, 96(6): 3228-33.
59. Mucke, L., Masliah, E., Yu, G.Q. et al. *High-level neuronal expression of abeta 1-42 in wild-type human amyloid protein precursor transgenic mice: Synaptotoxicity without plaque formation.* J Neurosci 2000**,** 20(11): 4050-8.
60. Oddo, S., Caccamo, A., Shepherd, D.H. et al. *Triple-transgenic model of Alzheimer's disease with plaques and tangles: Intracellular A[beta] and synaptic dysfunction.* Neuron 2003**,** 39(3): 409-21.
61. Lombardo, J.A., Stern, E.A., McLellan, M.E., Kajdasz, S.T., Hickey, G.A., Bacskai, B.J., Hyman, B.T. *Amyloid-beta antibody treatment leads to rapid normalization of plaque- induced neuritic alterations.* J Neurosci 2003, 23(34): 10879-83.
62. Carlsson, G., Ahlin, A., Dahllöf, G., Elinder, G., Henter, J.I., Palmblad, J. *Efficacy and safety of two different rG-CSF preparations in the treatment of patients with severe congenital neutropenia.* Br J Haematol 2004, 126(1): 127-32.
63. Kocherlakota, P., La Gamma, E.F. *Human granulocyte colony-stimulating factor may improve outcome attributable to neonatal sepsis complicated by neutropenia.* Pediatrics 1997, 100(1): E6.
64. Welte, K., Gabrilove, J., Bronchud, M.H., Platzer, E., Morstyn, G. *Filgrastim (r-metHuG-CSF): The first 10 years.* Blood 1996, 88(6): 1907-29.
65. Schabitz, W.R., Schneider, A. *Developing granulocyte-colony stimulating factor for the treatment of stroke: Current status of clinical trials.* Stroke 2006, 37(7): 1654; author reply 1655.
66. Takano, H., Ohtsuka, M., Akazawa, H. et al. *Pleiotropic effects of cytokines on acute myocardial infarction: G-CSF as a novel therapy for acute myocardial infarction.* Curr Pharm Des 2003, 9(14): 1121-7.
67. Kuethe, F., Figulla, H.R., Voth, M. et al. *[Mobilization of stem cells by granulocyte colony-stimulating factor for the regeneration of myocardial tissue after myocardial infarction].* Dtsch Med Wochenschr 2004, 129(9): 424-8.
68. Nienaber, C.A., Petzsch, M., Kleine, H.D., Eckard, H., Freund, M., Ince, H. *Effects of granulocyte-colony-stimulating factor on mobilization of bone-marrow-derived stem cells after myocardial infarction in humans.* Nat Clin Pract Cardiovasc Med 2006, 3(Suppl. 1): S73-7.
69. Ripa, R.S., Jørgensen, E., Wang, Y. et al. *Stem cell mobilization induced by subcutaneous granulocyte-colony stimulating factor to improve cardiac regeneration after acute ST-elevation myocardial infarction: Result of the double-blind, randomized, placebo-controlled stem cells in myocardial infarction (STEMMI) trial.* Circulation 2006, 113(16): 1983-92.
70. Suzuki, K., Nagashima, K., Arai, M. et al. *Effect of granulocyte colony-stimulating factor treatment at a low dose but for a long duration in patients with coronary heart disease.* Circ J 2006, 70(4): 430-7.
71. Sanchez-Ramos, J. *Clinical trial supported by AD Drug Discovery Foundation "Efficacy and Safety of Filgastrim as a Procognitive Agent in Alzheimer's Disease".* 2009.
72. de la Rubia, J., de Arriba, F., Arbona, C. et al. *Follow-up of healthy donors receiving granulocyte colony-stimulating factor for peripheral blood progenitor cell mobilization and collection. Results of the Spanish Donor Registry.* Haematologica 2008, 93(5): 735-40.
73. Stroncek, D.F., Dittmar, K., Shawker, T., Heatherman, A., Leitman, S.F. *Transient spleen enlargement in peripheral blood progenitor cell donors given G-CSF.* J Transl Med 2004, 2(1): 25.
74. Tigue, C.C., McKoy, K.M., Evens, A.M., Trifilio, S.M., Tallman, M.S., Bennett, C.L. *Granulocyte-colony stimulating factor administration to healthy individuals and persons with chronic neutropenia or cancer: An overview of safety considerations from the Research on Adverse Drug Events and Reports project.* Bone Marrow Transplant 2007 40(3): 185-92.
75. Broudy, V.C. *Stem cell factor and hematopoiesis.* Blood 1997, 90(4): 1345-64.
76. Hirota, S., Ito, A., Morii, E., Wanaka, A., Tohyama, M., Kitamura, Y., Nomura, S. *Localization of mRNA for c-kit receptor and its ligand in the brain of adult rats: An analysis using in situ hybridization histochemistry.* Mol Brain Res 1992, 15(1-2): 47-54.
77. Matsui, Y., Zsebo, K.M., Hogan, B.L.M. *Embryonic expression of a haematopoietic growth factor encoded by the SI locus and the ligand for c-kit.* Nature 1990, 347(6294): 667-9.
78. Morii, E., Hirota, S., Kim, H.M., Mikoshiba, K., Nishimune, Y., Kitamura, Y., Nomura, S. *Spatial expression of genes encoding c-kit receptors and their ligands in mouse cerebellum as revealed by in situ hybridization*. Brain Res Dev Brain Res 1992, 65(1): 123-6.
79. Akin, C., Metcalfe, D.D. *The biology of Kit in disease and the application of pharmacogenetics.* J Allergy Clin Immunol 2004, 114(1): 13-9; quiz 20.
80. Erlandsson, A., Larsson, J., Forsberg-Nilsson, K. *Stem cell factor is a chemoattractant and a survival factor for CNS stem cells.* Exp Cell Res 2004, 301(2): 201-10.
81. Jin, K., Mao, X.O., Sun, Y., Xie, L., Greenberg, D.A. *Stem cell factor stimulates neurogenesis in vitro and in vivo.* J Clin Invest 2002, 110(3): 311-9.
82. Dhandapani, K.M., Wade, F.M., Wakade, C., Mahesh, V.B., Brann, D.W. *Neuroprotection by stem cell factor in rat cortical neurons involves AKT and NFkappaB.* J Neurochem 2005, 95(1): 9-19.
83. Zhang, S.-C., Fedoroff, S. *Expression of stem cell factor and c-kit receptor in neural cells after brain injury.* Acta Neuropathol 1999, 97(4): 393-8.

84. Sun, L., Lee, J., Fine, H.A. *Neuronally expressed stem cell factor induces neural stem cell migration to areas of brain injury.* J Clin Invest 2004, 113(9): 1364-74.

85. Corti, S., Locatelli, F., Strazzer, S. et al. *Modulated generation of neuronal cells from bone marrow by expansion and mobilization of circulating stem cells with in vivo cytokine treatment.* Exp Neurol 2002, 177(2): 443-52.

86. Zhao, L.-R., Navalitloha, Y., Singhal, S. et al. *Hematopoietic growth factors pass through the blood-brain barrier in intact rats.* Exp Neurol 2007, 204(2): 569-73.

87. Leyhe, T., Hoffmann, N., Stransky, E., Laske, C. *Increase of SCF plasma concentration during donepezil treatment of patients with early Alzheimer's disease.* Int J Neuropsychopharmacol 2009, 12(10): 1319-26.

88. Zhang, S.-C., Fedoroff, S. *Modulation of microglia by stem cell factor.* J Neurosci Res 1998, 53(1): 29-37.

89. Katafuchi, T., Li, A.J., Hirota, S., Kitamura, Y., Hori, T. *Impairment of spatial learning and hippocampal synaptic potentiation in c-kit mutant rats.* Learning Memory 2000, 7(6): 383-92.

90. Laske, C., Stellos, K., Stransky, E. et al. *Decreased plasma and cerebrospinal fluid levels of stem cell factor in patients with early Alzheimer's disease.* J Alzheimer's Dis 2008, 15(3): 451-60.

91. Miyazawa, K., Williams, D.A., Gotoh, A., Nishimaki, J., Broxmeyer, H.E., Toyama, K. *Membrane-bound Steel factor induces more persistent tyrosine kinase activation and longer life span of c-kit gene-encoded protein than its soluble form.* Blood 1995, 85(3): 641-9.

92. Piao, C.-S., Gonzalez-Toledo, M.E., Xue, Y.Q. et al. *The role of stem cell factor and granulocyte-colony stimulating factor in brain repair during chronic stroke.* J Cereb Blood Flow Metab 2009, 29(4): 759-70.

93. Nicola, N.A. *GM-CSF*. In: Cytokine Reference. Oppenheim, J.J., Feldmann, M. (Eds.). San Diego: Academic Press, 2001.

94. Koguchi, K., Nakatsuji, Y., Okuno, T., Sawada, M., Sakoda, S. *Microglial cell cycle-associated proteins control microglial proliferation in vivo and in vitro and are regulated by GM-CSF and density-dependent inhibition.* J Neurosci Res 2003, 74(6): 898-905.

95. Schabitz, W.R., Krüger, C., Pitzer, C. et al. *A neuroprotective function for the hematopoietic protein granulocyte-macrophage colony stimulating factor (GM-CSF).* J Cereb Blood Flow Metab 2008, 28(1): 29-43.

96. Maurer, M.H., Schäbitz, W.R., Schneider, A. *Old friends in new constellations - The hematopoetic growth factors G-CSF, GM-CSF, and EPO for the treatment of neurological diseases.* Curr Med Chem 2008, 15(14): 1407-11.

97. Sawada, M., Itoh, Y., Suzumura, A., Marunouchi, T. *Expression of cytokine receptors in cultured neuronal and glial cells.* Neurosci Lett 1993, 160(2): 131-4.

98. Guillemin, G., Boussin, F.D., Le Grand, R., Croitoru, J., Coffigny, H., Dormont, D. *Granulocyte macrophage colony stimulating factor stimulates in vitro proliferation of astrocytes derived from simian mature brains.* Glia 1996, 16(1): 71-80.

99. Kruger, C., Laage, R., Pitzer, C., Schäbitz, W.R. Schneider, A. *The hematopoietic factor GM-CSF (granulocyte-macrophage colony-stimulating factor) promotes neuronal differentiation of adult neural stem cells in vitro.* BMC Neurosci 2007, 8: 88.

100. Mirski, R., Reichert, F., Klar, A., Rotshenker, S. *Granulocyte macrophage colony stimulating factor (GM-CSF) activity is regulated by a GM-CSF binding molecule in Wallerian degeneration following injury to peripheral nerve axons.* J Neuroimmunol 2003, 140(1-2): 88-96.

101. Reichert, F., Saada, A., Rotshenker, S. *Peripheral nerve injury induces Schwann cells to express two macrophage phenotypes: Phagocytosis and the galactose-specific lectin MAC-2.* J Neurosci 1994, 14(5): 3231-45.

102. Schermer, C., Humpel, C. *Granulocyte macrophage-colony stimulating factor activates microglia in rat cortex organotypic brain slices.* Neurosci Lett 2002, 328(2): 180-4.

103. Suzumura, A., Sawada, M., Marunouchi, T. *Selective induction of interleukin-6 in mouse microglia by granulocyte-macrophage colony-stimulating factor.* Brain Res 1996, 713(1-2): 192-8.

104. Coxon, A., Tang, T., Mayadas, T.N. *Cytokine-activated endothelial cells delay neutrophil apoptosis in vitro and in vivo: A role for granulocyte/macrophage colony-stimulating factor.* J Exp Med 1999, 190(7): 923-34.

105. Ha, Y. *Synthes Award for Resident Research on Spinal Cord and Spinal Column Injury: Granulocyte macrophage colony stimulating factor (GM-CSF) prevents apoptosis and improves functional outcome in experimental spinal cord contusion injury.* Clin Neurosurg 2005, 52: 341-7.

106. Manczak, M., Mao, P., Nakamura, K., Bebbington, C., Park, B., Reddy, P.H. *Neutralization of granulocyte macrophage colony stimulating factor decreases amyloid beta 1-42 and suppresses microglial activity in a transgenic mouse model of Alzheimer's disease.* Hum Mol Genet 2009, 18(20): 3876-93.

107. Tarkowski, E., Wallin, A., Regland, B., Blennow, K., Tarkowski, A. *Local and systemic GM-CSF increase in Alzheimer's disease and vascular dementia.* Acta Neurol Scand 2001, 103(3): 166-74.

108. Kim, H.-D., Kong, F.K., Cao, Y., Lewis, T.L., Kim, H., Tang, D.C., Fukuchi, K. *Immunization of Alzheimer model mice with adenovirus vectors encoding amyloid [beta]-protein and GM-CSF reduces amyloid load in the brain.* Neurosci Lett 2004, 370(2-3): 218-23.

109. DaSilva, K.A., Brown, M.E., Westaway, D., McLaurin, J. *Immunization with amyloid-[beta] using GM-CSF and IL-4 reduces amyloid burden and alters plaque morphology.* Neurobiol Dis 2006, 23(2): 433-44.

110. Tarkowski, E., Rosengren, L., Blomstrand, C., Wikkelsö, C., Jensen, C., Ekholm, S., Tarkowski, A. *Intrathecal release of pro- and anti-inflammatory cytokines during stroke.* Clin Exp Immunol 1997, 110(3): 492-9.

111. Tarkowski, E., Andreasen, N., Tarkowski, A., Blennow, K. *Intrathecal inflammation precedes development of Alzheimer's disease.* J Neurol Neurosurg Psychiatry 2003, 74(9): 1200-5.

112. Stanley, E.R. *Colony stimulating factor-1 (macrophage colony stimulating factor).* In: The Cytokine Handbook. Thomson, A.W. (Ed.). San Diego: Academic Press, 1994, 387-418.

113. Giulian, D., Ingeman, J.E. *Colony-stimulating factors as promoters of ameboid microglia.* J Neurosci 1988, 8(12): 4707-17.

114. Boissonneault, V., Filali, M., Lessard, M., Relton, J., Wong, G., Rivest, S. *Powerful beneficial effects of macrophage colony-stimulating factor on beta-amyloid deposition and cognitive impairment in Alzheimer's disease.* Brain 2009, 132(Pt. 4): 1078-92.

115. Davoust, N., Vuaillat, C., Cavillon, G. et al. *Bone marrow CD34+/B220+ progenitors target the inflamed brain and display in vitro differentiation potential toward microglia.* FASEB J 2006, 20(12): 2081-92.

116. Raivich, G., Moreno-Flores, M.T., Möller, J.C., Kreutzberg, G.W. *Inhibition of posttraumatic microglial proliferation in a genetic model of macrophage colony-stimulating factor deficiency in the mouse.* Eur J Neurosci 1994, 6(10): 1615-8.

117. Vidyadaran, S., Ooi, Y.Y., Subramaiam, H., Badiei, A., Abdullah, M., Ramasamy, R., Seow, H.F. *Effects of macrophage colony-stimulating factor on microglial responses to lipopolysaccharide and beta amyloid.* Cell Immunol 2009, 259(1): 105-10.

118. Ray, S., Britschgi, M., Herbert, C. et al. *Classification and prediction of clinical Alzheimer's diagnosis based on plasma signaling proteins.* Nat Med 2007, 13(11): 1359-62.

119. Lapidot, T., Dar, A., Kollet, O. *How do stem cells find their way home?* Blood 2005, 106(6): 1901-10.

120. Zhou, B., Han, Z.C., Poon, M.C., Pu, W. *Mesenchymal stem/stromal cells (MSC) transfected with stromal derived factor 1 (SDF-1) for therapeutic neovascularization: Enhancement of cell recruitment and entrapment.* Med Hypotheses 2007, 68(6): 1268-71.

121. Aiuti, A., Webb, I.J., Bleul, C., Springer, T., Gutierrez-Ramos, J.C. *The chemokine SDF-1 is a chemoattractant for human CD34+ hematopoietic progenitor cells and provides a new mechanism to explain the mobilization of CD34+ progenitors to peripheral blood.* J Exp Med 1997, 185(1): 111-20.

122. Bleul, C.C., Fuhlbrigge, R.C., Casasnovas, J.M., Aiuti, A., Springer, T.A. *A highly efficacious lymphocyte chemoattractant, stromal cell-derived factor 1 (SDF-1).* J Exp Med 1996, 184(3): 1101-9.

123. Carr, A.N., Howard, B.W., Yang, H.T. et al. *Efficacy of systemic administration of SDF-1 in a model of vascular insufficiency: Support for an endothelium-dependent mechanism.* Cardiovasc Res 2006, 69(4): 925-35.

124. Langer, H., May, A.E., Daub, K. et al. *Adherent platelets recruit and induce differentiation of murine embryonic endothelial progenitor cells to mature endothelial cells in vitro.* Circ Res 2006, 98(2): e2-10.

125. Stellos, K., Langer, H., Daub, K. et al. *Platelet-derived stromal cell-derived factor-1 regulates adhesion and promotes differentiation of human CD34+ cells to endothelial progenitor cells.* Circulation 2008, 117(2): 206-15.

126. Robin, A.M., Zhang, Z.G., Wang, L. et al. *Stromal cell-derived factor 1alpha mediates neural progenitor cell motility after focal cerebral ischemia.* J Cereb Blood Flow Metab 2006, 26(1): 125-34.

127. Stumm, R., Hollt, V. *CXC chemokine receptor 4 regulates neuronal migration and axonal pathfinding in the developing nervous system: Implications for neuronal regeneration in the adult brain.* J Mol Endocrinol 2007, 38(3): 377-82.

128. Laske, C., Stellos, K., Eschweiler, G.W., Leyhe, T., Gawaz, M. *Decreased CXCL12 (SDF-1) plasma levels in early Alzheimer's disease: A contribution to a deficient hematopoietic brain support?* J Alzheimers Dis 2008, 15(1): 83-95.

129. Maiese, K., Li, F., Chong, Z.Z. *Erythropoietin in the brain: Can the promise to protect be fulfilled?* Trends Pharmacol Sci 2004, 25(11): 577-83.

130. Maiese, K., Li, F., Chong, Z.Z. *New avenues of exploration for erythropoietin.* JAMA 2005, 293(1): 90-5.

131. Chong, Z.Z., Kang, J.Q., Maiese, K. *Hematopoietic factor erythropoietin fosters neuroprotection through novel signal transduction cascades.* J Cereb Blood Flow Metab 2002, 22(5): 503-14.

132. Fliser, D., Haller, H. *Erythropoietin and treatment of non-anemic conditions—cardiovascular protection.* Semin Hematol 2007, 44(3): 212-7.

133. Jacobson, L.O., Goldwasser, E., Fried, W., Plzak, L. *Role of the kidney in erythropoiesis.* Nature 1957, 179(4560): 633-4.

134. Lasne, F., de Ceaurriz, J. *Recombinant erythropoietin in urine.* Nature 2000, 405(6787): 635.

135. Sathyanarayana, P., Menon, M.P., Bogacheva, O. et al. *Erythropoietin modulation of podocalyxin and a proposed erythroblast niche.* Blood 2007, 110(2): 509-18.

136. Schmeding, M., Neumann, U.P., Boas-Knoop, S., Spinelli, A., Neuhaus, P. *Erythropoietin reduces ischemia-reperfusion injury in the rat liver.* Eur Surg Res 2007, 39(3): 189-97.

137. Fisher, J.W. *Erythropoietin: Physiology and pharmacology update.* Exp Biol Med (Maywood) 2003, 228(1): 1-14.

138. Brines, M.L., Ghezzi, P., Keenan, S. et al. *Erythropoietin crosses the blood-brain barrier to protect against experimental brain injury.* Proc Natl Acad Sci U S A 2000, 97(19): 10526-31.

139. Juul, S.E., Yachnis, A.T., Rojiani, A.M., Christensen, R.D. *Immunohistochemical localization of erythropoietin and its receptor in the developing human brain.* Pediatr Dev Pathol 1999, 2(2): 148-58.

140. Marti, H.H., Wenger, R.H., Rivas, L.A. *Erythropoietin gene expression in human, monkey and murine brain.* Eur J Neurosci 1996, 8(4): 666-76.

141. Morishita, E., Masuda, S., Nagao, M., Yasuda, Y., Sasaki, R. *Erythropoietin receptor is expressed in rat hippocampal and cerebral cortical neurons, and erythropoietin prevents in vitro glutamate-induced neuronal death.* Neuroscience 1997, 76(1): 105-16.

142. Sairanen, T., Karjalainen-Lindsberg, M.L., Paetau, A., Ijäs, P., Lindsberg, P.J. *Apoptosis dominant in the periinfarct area of human ischaemic stroke–A possible target of antiapoptotic treatments.* Brain 2006, 129(Pt. 1): 189-99.

143. Sirén, A.L., Knerlich, F., Poser, W., Gleiter, C.H., Brück, W., Ehrenreich, H. *Erythropoietin and erythropoietin receptor in human ischemic/hypoxic brain.* Acta Neuropathol 2001, 101(3): 271-6.

144. Juul, S.E., Stallings, S.A., Christensen, R.D. *Erythropoietin in the cerebrospinal fluid of neonates who sustained CNS injury.* Pediatr Res 1999, 46(5): 543-7.

145. Springborg, J.B., Sonne, B., Frederiksen, H.J. et al. *Erythropoietin in the cerebrospinal fluid of patients with aneurysmal subarachnoid haemorrhage originates from the brain.* Brain Res 2003, 984(1-2): 143-8.

146. Assaraf, M.I., Diaz, Z., Liberman, A. et al. *Brain erythropoietin receptor expression in Alzheimer disease and mild cognitive impairment.* J Neuropathol Exp Neurol 2007, 66(5): 389-98.

147. Barkats, M., Millecamps, S., Abrioux, P., Geoffroy, M.C., Mallet, J. *Overexpression of glutathione peroxidase increases the resistance of neuronal cells to Abeta-mediated neurotoxicity.* J Neurochem 2000, 75(4): 1438-46.

148. Boldogh, I., Kruzel, M.L. *Colostrinin: An oxidative stress modulator for prevention and treatment of age-related disorders.* J Alzheimers Dis 2008, 13(3): 303-21.

149. Moon, J.H., Kim, S.Y., Lee, H.G., Kim, S.U., Lee, Y.B. *Activation of nicotinic acetylcholine receptor prevents the production of reactive oxygen species in fibrillar beta amyloid peptide (1-42)-stimulated microglia.* Exp Mol Med 2008, 40(1): 11-8.

150. Sultana, R., Newman, S., Mohmmad-Abdul, H., Keller, J.N., Butterfield, D.A. *Protective effect of the xanthate, D609, on Alzheimer's amyloid beta-peptide (1-42)-induced oxidative stress in primary neuronal cells.* Free Radic Res 2004, 38(5): 449-58.

151. Li, G., Ma, R., Huang, C. et al. *Protective effect of erythropoietin on beta-amyloid-induced PC12 cell death through antioxidant mechanisms.* Neurosci Lett 2008, 442(2): 143-7.

152. Ma, R., Ma, R., Huang, C. *Erythropoietin protects PC12 cells from beta-amyloid(25-35)-induced apoptosis via PI3K/Akt signaling pathway.* Neuropharmacology 2009, 56(6-7): 1027-34.

153. Bornemann, K.D., Wiederhold, K.H., Paupli, C. et al. *Abeta-induced inflammatory processes in microglia cells of APP23 transgenic mice.* Am J Pathol 2001, 158(1): 63-73.

154. Chong, Z.Z., Li, F., Maiese, K. *Oxidative stress in the brain: Novel cellular targets that govern survival during neurodegenerative disease.* Prog Neurobiol 2005, 75(3): 207-46.

155. Chong, Z.Z., Li, F., Maiese, M. *Activating Akt and the brain's resources to drive cellular survival and prevent inflammatory injury.* Histol Histopathol 2005, 20(1): 299-315.

156. Kang, J.Q., Chong, Z.Z., Maiese, K. *Akt1 protects against inflammatory microglial activation through maintenance of membrane asymmetry and modulation of cysteine protease activity.* J Neurosci Res 2003, 74(1): 37-51.

157. Kang, J.Q., Chong, Z.Z., Maiese, K. *Critical role for Akt1 in the modulation of apoptotic phosphatidylserine exposure and microglial activation.* Mol Pharmacol 2003, 64(3): 557-69.

158. Wang, C.Y., Mayo, M.W., Korneluk, R.G., Goeddel, D.V., Baldwin, A.S. Jr. *NF-kappaB antiapoptosis: Induction of TRAF1 and TRAF2 and c-IAP1 and c- IAP2 to suppress caspase-8 activation.* Science 1998, 281(5383): 1680-3.

159. Reed, J.C. *Apoptosis-regulating proteins as targets for drug discovery.* Trends Mol Med 2001, 7(7): 314-9.

160. Chong, Z.Z., Li, F., Maiese, K. *Erythropoietin requires NF-kappaB and its nuclear translocation to prevent early and late apoptotic neuronal injury during beta-amyloid toxicity.* Curr Neurovasc Res 2005, 2(5): 387-99.

161. Jamsa, A., Hasslund, K., Cowburn, R.F., Bäckström, A., Vasänge, M. *The retinoic acid and brain-derived neurotrophic factor differentiated SH-SY5Y cell line as a model for Alzheimer's disease-like tau phosphorylation.* Biochem Biophys Res Commun 2004, 319(3): 993-1000.

162. Ishizawa, T., Sahara, N., Ishiguro, K. et al. *Co-localization of glycogen synthase kinase-3 with neurofibrillary tangles and granulovacuolar degeneration in transgenic mice.* Am J Pathol 2003, 163(3): 1057-67.

163. Sun, Z.K., Yang, H.Q., Pan, J., Zhen, H., Wang, Z.Q., Chen, S.D., Ding, J.Q. *Protective effects of erythropoietin on tau phosphorylation induced by beta- amyloid.* J Neurosci Res 2008, 86(13): 3018-27.

164. Danielyan, L., Schäfer, R., Schulz, A. et al. *Survival, neuron-like differentiation and functionality of mesenchymal stem cells in neurotoxic environment: The critical role of erythropoietin.* Cell Death Differ 2009, 16(12): 1599-614.

165. Zwezdaryk, K.J., Coffelt, S.B., Figueroa, Y.G. et al. *Erythropoietin, a hypoxia-regulated factor, elicits a pro-angiogenic program in human mesenchymal stem cells.* Exp Hematol 2007, 35(4): 640-52.

166. Tsang, S.W., Pomakian, J., Marshall, G.A. et al. *Disrupted muscarinic M1 receptor signaling correlates with loss of protein kinase C activity and glutamatergic deficit in Alzheimer's disease.* Neurobiol Aging 2007, 28(9): 1381-7.

167. Robinson, S.R. *Neuronal expression of glutamine synthetase in Alzheimer's disease indicates a profound impairment of metabolic interactions with astrocytes.* Neurochem Int 2000, 36(4-5): 471-82.

168. de Hemptinne, I., Vermeiren, C., Maloteaux, J.M., Hermans, E. *Induction of glial glutamate transporters in adult mesenchymal stem cells.* J Neurochem 2004, 91(1): 155-66.