Psychiatr. Pol. 2014; 48(6):1073–1085
PL ISSN 0033-2674
www.psychiatriapolska.pl

Stem cell research and its growing impact on contemporary psychiatry

Mariusz Z. Ratajczak[1,2], Jolanta Kucharska-Mazur[3], Jerzy Samochowiec[3]

[1]Stem Cell Institute, James Graham Brown Cancer Center, University of Louisville, Louisville, Kentucky, USA
Head: prof. dr hab. n. med. M. Z. Ratajczak

[2]Department of Physiology Pomeranian Medical University, Szczecin, Poland
Head: prof. dr hab. n. med. M. Z. Ratajczak

[3] Department of Psychiatry Pomeranian Medical University, Szczecin, Poland
Head: prof. dr hab. n. med. J. Samochowiec

Summary

The expanding field of stem cell research is now beginning to help with the problems of modern psychiatry. On the one hand, induced pluripotent stem cells (iPSCs) can now be used to generate neural cell lines from patients suffering from psychiatric disorders, which can then serve as models for studying changes in gene expression pattern involved in the pathogenesis of these diseases. These artificially generated neural cells are also employed in studying the efficacy of newly developed antipsychotic treatments. On the other hand, evidence has accumulated that not only monocytes, which can be microglia precursors, but also certain other adult bone marrow-derived cells may cross the blood–brain barrier and affect biological processes in brain tissue. Along with evidence of circulating and brain-infiltrating cells, there are well-studied factors (e.g., chemokines, phosphosphingolipids, and complement-cleavage fragments) that modulate trafficking of these cells between bone marrow and neural tissue. These observations may help to shed new light on the pathogenesis of psychotic disorders and, in the future, perhaps help to develop more effective treatments.

Key words: circulating stem cells, VSEL, sphingosine-1-phosphate

Acknowledgment: This work was supported by EU structural funds, the Innovative Economy Operational Program POIG.01.01.01-00-109/09-01, and Maestro grant 2011/02/A/NZ4/00035 to MZR.

Introduction

Rapid progress in stem cell research touches all of medical specialties, and psychiatry is no exception. This is understandable, because development and even the postnatal state of brain tissue and its function are affected by circulating cells, including stem cells [1-3]. An important step forward in studying psychiatric disorders is based on the possibility of obtaining patient-specific neural cell lines derived from induced pluripotent stem cells (iPSCs) created from patient's own somatic cells, such as skin cells [4]. Such neural cell lines allow the study of changes in the gene expression patterns involved in psychiatric syndromes and the testing of novel treatment strategies.

Moreover, evidence indicates that some bone marrow (BM)-derived cells, such as precursors of microglia, can cross the blood–brain barrier not only in situations when this barrier is damaged, as seen for example in neurodegenerative disorders, but also under steady-state conditions [5].

It is well known that peripheral blood (PB) serves as a "highway" for circulating stem cells. Hematopoietic stem cells (HSCs), endothelial progenitors (EPCs), mesenchymal stem cells (MSCs), and developmentally early very small embryonic-like stem cells (VSELs) circulate in PB at low concentrations, and their number increases in several models of tissue and organ damage, including models of acute myocardial infarction, stroke, skin burns, acute intestinal inflammatory diseases, and, recently, psychotic syndromes [1-3, 6]. Therefore, increase in the number of these cells in PB may have diagnostic and prognostic value.

Trafficking of stem cells in PB is triggered and governed by several factors, including activation of the complement and coagulation cascades and increases in the levels of some chemokines, cytokines, growth factors, bioactive phosphosphingolipids, and even extracellular nucleotides [7].

In this review we will focus on these novel developments in stem cell research that are becoming increasingly relevant to understanding the pathogenesis of psychotic disorders.

Induced pluripotent stem cells (iPSCs) as tools to study psychotic disorders and test new treatment strategies

The development of induced pluripotent stem cell (iPSC) technology allows one to obtain customized cell lines derived from cells isolated from a given patient [4]. These ethically non-controversial stem cells are derived by genetic modification of mature postnatal somatic cells (Figure 1) by their transformation *in vitro* using genes encoding key transcription factors involved in the early stages of embryogenesis (i.e., Oct-4, Nanog, Klf4, and c-myc). These genes regulating embryonic development are introduced into somatic cells (e.g., skin fibroblasts) using retroviral vectors [4]. As a result of this strategy, transformed iPSCs that differentiate into cells derived from all three germ layers (meso-, ecto-, and endoderm) can be obtained. Such transformation is, however, relatively rare, as, on average, only one cell in several thousand undergoing the aforementioned genetic manipulation is transformed (induced to the embryonic

stage) and begins to proliferate, creating a clone consisting of iPSCs [4]. Recently, some modifications to this strategy, that employ more limited number of genes in the transduction process, microRNAs (miRNAs), or even small molecules that modify DNA structure, have been described [4]. iPSCs have been proposed as an ethically acceptable source of pluripotent stem cells that are an alternative to stem cells isolated from embryos (ESCs). What is highly relevant for psychotic disorders, such iPSCs can be generated from somatic cells isolated from the patient and differentiated into neural cells as models to study changes in gene expression of cells affected by schizophrenia. This interesting tool could help to identify new genetic lesions, and such cells could also be employed to test novel treatment strategies.

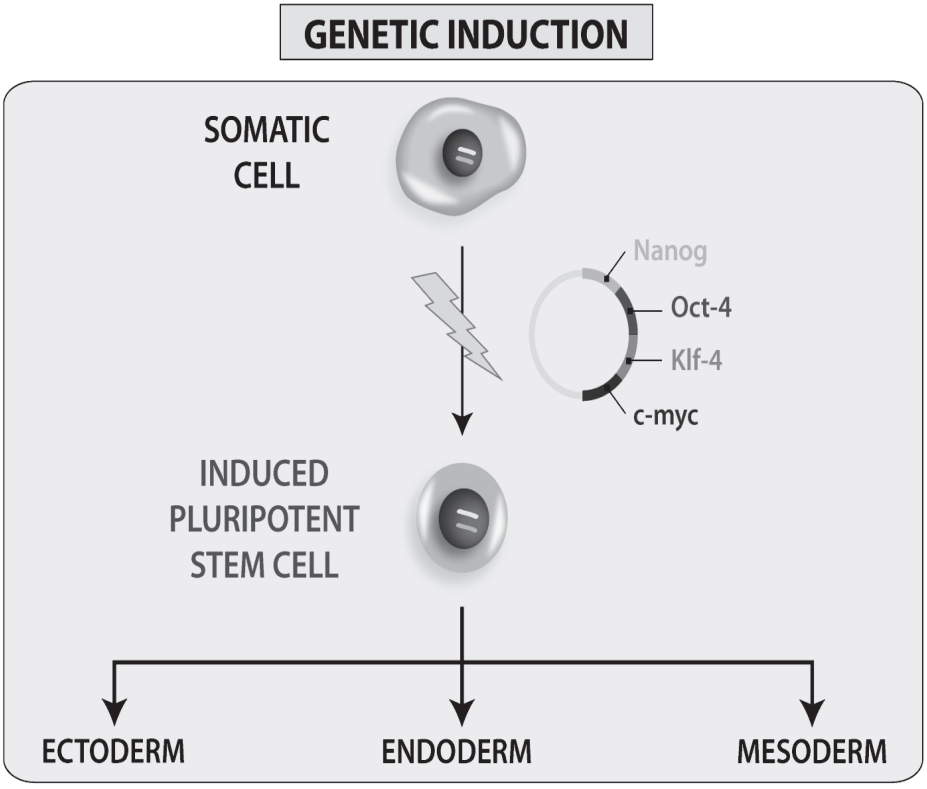

Figure 1. **Strategy to generate induced pluripotent stem cells (iPSCs) from somatic cells.** Normal patient-derived somatic cells (for example skin fibroblasts) are reprogrammed (transformed) into immortalized pluripotent stem cells using a mixture of genes that encode embryonic transcription factors (e.g., Oct-4, Nanog, Klf-4, and c-myc). Such iPSCs can be further differentiated into neural cells (derived from ectoderm) or cells derived from the other two primary germ cell layers.

Stem cells circulating in peripheral blood and their relevance to pathological states

As mentioned above, PB is a "highway" for circulating stem cells, and it is well known that HSCs are non-stop travellers throughout the body in both time and space [8]. They circulate in PB and lymph during development, moving between major anatomical sites where hematopoiesis is initiated and/or temporarily active. Starting from the blood islands in the yolk sac of the embryo, HSCs move through the aortic endothelium, placental vessels, and spleen until they reach the fetal liver in the second trimester of gestation. By the third trimester of gestation, they reach their final destination, i.e. the BM microenvironment, where they reside in adult life.

In addition to HSCs, other types of stem cells may also circulate in PB, even under normal steady-state conditions. The most numerous among circulating stem cells are, of course, HSCs, which use this route in postnatal life to keep the pool of stem cells in the BM of bones located in different areas of the body in balance. Moreover, during infection, circulating HSCs may differentiate directly in damaged tissues into progenitor cells of the myeloid lineage and locally supply granulocytes, monocytes, and dendritic cells to fight the infection [9].

As mentioned above, in addition to HSCs there are also MSCs, EPCs, and VSELs detectable in PB. It has been proposed that these different types of cells circulate under steady-state conditions in order to "patrol" peripheral tissues for potential injuries and may play the role of "paramedics" in repairing minor lesions in peripheral organs [3].

Moreover, these circulating cells have been demonstrated in PB in several models of more serious tissue organ injury, including models of heart infarct, stroke, skin burns, and intestinal inflammation, and our team has also recently reported their presence in PB in psychotic disorders [3, 6, 10-12]. Therefore, the number of these cells detected in PB may be of both diagnostic and prognostic value.

Overall, the number of stem cells circulating in PB increases in response to 1) systemic or local inflammation, 2) strenuous exercise, 3) tissue or organ injury, and 4) pharmacological agents [8, 9]. Specifically, after administration of certain agents that induce their forced egress into PB, a process known as stem cell mobilization, the number of HSCs circulating in PB may increase up to 100-fold. Pharmacological mobilization has been exploited in hematological transplantology as a mean to obtain HSPCs for hematopoietic reconstitution. The most important mobilizing agents currently employed in the clinical practice are granulocyte colony stimulating factor (G-CSF) and CXCR4 receptor-blocking molecules (e.g., AMD3100) [13, 14].

Relevant for this review, lithium, a drug employed for over 50 years in psychiatry to treat mood instability, has also been described as inducing stem cell mobilization, increasing the plasma level of G-CSF, and potentiating the mobilizing effects of G-CSF [15].

Endogenous factors increasing the number of circulating stem cells in PB

Stem cells are located in the BM and other organs in areas known as stem cell niches. In BM, for example, the major role in retention of HSCs is played by the chemokine stromal-derived factor 1 (SDF-1)–CXCR4 receptor axis [16]. While SDF-1 is expressed by cells within the niche, CXCR4 is present on the surface of HSCs. Stem cell niches are present also in brain, and neuronal stem cells have been described as residing in the subventricular zone of the lateral ventricles and olfactory bulb, as well as in the subgranular zone of the dentate gyrus in the hippocampus [17-19]. The factors responsible for retention of these cells in brain stem cell niches are still not very well understood. There are, however, indications that the SDF-1–CXCR4 axis may play a role there as well [20-21].

Overall, the egress of HSCs from BM into PB during the mobilization process has been proposed to be directed by a decrease in the SDF-1–CXCR4 interaction in BM niches (e.g., due to release of proteolytic enzymes from granulocytes, which degrades SDF-1) and an increase in the PB level of SDF-1, which would reverse the trans-endothelial chemotactic gradient of SDF-1 between the BM microenvironment and plasma [14]. This concept, however, has been recently challenged by the observation that the PB SDF-1 level does not increase significantly during mobilization, and thus one cannot explain the egress of HSCs from BM to PB simply by the reversal of the gradient between BM and PB [22].

Recent research from our laboratories and confirmed also by others has established that sphingosine-1-phosphate (S1P) is a major chemoattractant for HSCs and is present in PB [22, 23]. A high concentration of S1P in PB plasma (and therefore a considerable chemotactic gradient with respect to BM) is continuously present under steady-state conditions. On the other hand, S1P may also play an important role in homing of cells to brain tissue. It is known that inhibitors of S1P-mediated migration (e.g., fingolimod) are employed to decrease infiltration of the brain by lymphocytes in multiple sclerosis patients [24]. Moreover, it has also been proposed that the release of neurotransmitters from the synapses of nerves that innervate the BM microenvironement (e.g., involving the dopamine, β2- and β3-adrenergic receptors) may affect stem cell mobilization [25], and this issue will be discussed latter in this review.

Evidence has accumulated that mobilization of stem cells is triggered by activation of the complement cascade in the BM microenvironment. In support of this mechanism, it has been observed that mice that do not activate the distal part of the complement cascade display a profound defect in mobilization of HSCs [26]. Moreover, the complement cascade becomes activated in all mechanisms leading to mobilization of HSCs (i.e., systemic inflammation, organ injury, and administration of all mobilizing drugs). As we recently demonstrated, the mobilization process is attenuated by heme oxygenase 1 (HO-1), which has anti-inflammatory and inhibitory effects and inhibits activation of the complement cascade [27].

Thus, it is important while investigating stem cell trafficking to also measure, in parallel, changes in S1P and SDF-1 levels in PB, as well as activation of the com-

plement cascade by detecting plasma levels of complement cleavage fragments (e.g., C3a, C5a, C5b-C9).

Trafficking of stem cells in psychiatric disorders

Since stem cells are mobilized in PB in several clinical situations, our group became interested in changes in the number of these circulating cells in psychiatric disorders. To address this question, we evaluated the stem cells circulating in PB, first in patients with anxiety disorders and then in patients with acute psychotic syndromes. We focused on the number of HSCs circulating in PB and subsequently the number of VSELs. The main idea behind this study was that circulating cells, including stem cells, could be involved in the pathogenesis of certain psychiatric disorders.

This hypothesis seems reasonable, as it has previously been proposed that release of neurotransmitters from the synapses of nerves that innervate the BM microenvironment (e.g., involving the dopamine, β2- and β3-adrenergic receptors) and enhanced tonus of the vegetative nervous system are responsible for the mobilization of HSCs into PB [25]. In support of this possibility, UDP-galactose:ceramide galactosyltransferase-deficient mice, which exhibit aberrant nerve conduction and do not release norepinephrine (NE) into the BM microenvironment, were found not to mobilize HSCs in response to administration of G-CSF [28]. By contrast, as recently reported, modification of sympathetic output does not affect G-CSF-induced mobilization in humans, as would be predicted. Specifically, normal human HSPC volunteer donors who were receiving NE reuptake inhibitors (NRI) because of depression or were treated with β2-blockers because of hypertension mobilize HSCs into PB in a similar manner as normal controls [29]. Surprisingly, mobilization in these patients was neither enhanced by NRI administration nor suppressed by β2-blockers, as one would expect based on the murine data reported in the literature.

Circulation of hematopoietic stem cells (HSCs) in patients with psychotic disorders and acute anxiety syndromes

To address this intriguing issue and the discrepancy between human and mice, we analyzed the levels of circulating HSCs in patients suffering from acute psychosis and anxiety disorders -clinical situations with elevated levels of catecholamine in PB [30]. It is well known that the levels of NE and dopamine are elevated in peripheral tissues and PB in these patients. Moreover, during acute psychotic syndromes, patients are under the influence of several neural mediators [30]. Moreover, one group reported a higher NE turnover rate in first-episode schizophrenic patients [31].

In our study we enrolled 30 unrelated individuals with a diagnosis of first-episode psychosis, which was assessed using The International Classification of Diseases 10th Revision criteria (ICD-10, 1998) [32]. Individuals enrolled in this study were diagnosed with first-episode psychosis (F20, F22, F31 or F23) according to ICD-10, with no history of axis I psychiatric disorders other than the above mentioned (drug-naïve) psychosis. Psychometric evaluation of patients was performed with the positive

and negative syndrome scale (PANSS) [33]. We also performed a study of 30 patients suffering from acute anxiety disorders (manuscript in preparation). All these patients were compared with an ethnic- and gender-matched control group of 35 healthy volunteers without psychiatric disorders, which were excluded according to an examination by a specialist psychiatrist. Patients with a history of serious lifetime medical events, organic brain injuries, or drug or alcohol dependence were excluded from the study.

Mobilization of HSCs in patients with first-episode psychosis, patients suffering from acute anxiety disorders, and control individuals was evaluated using 1) FACS (fluorescence-activated cell storting) to evaluate the number of HSCs circulating in PB identified by phenotypic staining as $CD34^+$, $CD133^+$, $CD34^+CD45^+Lin^-$ or $CD133^+CD45^+Lin^-$ cells and 2) functional *in vitro* assays to detect the number of granulo-monocytic (CFU-GM) and erythroid (BFU-E) clonogenic progenitors. In parallel, we measured the levels of epinephrine, norepinephrine (NE), and dopamine in PB serum. Both cells and catecholamine levels were enumerated in acute patients with psychotic and anxiety disorders before and after treatment and compared with age- and sex-matched controls.

In the performed studies, we did not observe any significant differences in the number of circulating HSCs or clonogenic BFU-E and CFU-GM progenitors between normal controls, and psychotic patients or patients with anxiety disorders [34]. In particular, the number of HSPCs circulating in PB was not affected by increased levels of adrenaline, norepinephrine, and dopamine in the PB of patients suffering from acute psychotic syndromes. Interestingly, the levels of NE and dopamine in our study were lower in patients with anxiety disorders and increased after treatment. Thus, our data argue against an effect of vegetative nervous system tone on the number of HSCs circulating in PB in humans. Our negative results, performed on patients suffering from acute psychoses and anxiety disorders, somewhat corroborates the results reported for normal HSPC volunteer donors that were previously treated with NRI because of depression or with β2-blockers because of high blood pressure and mobilized with G-CSF [29]. This finding suggests that there are some clear differences between rodents and humans in the effect of the vegetative nervous system on mobilization of HSCs.

Circulation of very small embryonic-like stem cells (VSELs) in patients with psychotic disorders and acute anxiety syndromes

Next, we tested whether VSELs and factors that modulate their trafficking may be biological markers for acute psychosis and anxiety syndrome [6]. To address this question, 28 subjects during their first non-affective psychotic episode were investigated before and after antipsychotic treatment and were compared with 35 healthy controls (CG); the psychotic group (PG) was divided into "schizophrenic" (SG) and "non-schizophrenic" (NG) subgroups. We also investigated 30 patients suffering from acute anxiety disorders. In all patients we employed FACS analysis to determine the number of $Lin^-/CD45^-/CD34^+$ and $Lin^-/CD45^-/CD133^+$ very small embryonic-like stem cells (VSELs) circulating in PB, which express markers of early-development stem cells

and neural lineage markers such as Oct-4, Sox2, Nanog, GFAP, Olig1, Olig2, Musashi, Nestin, and βIII-tubulin.

We found that the mean number of $Lin^-/CD45^-/CD34^+$ VSELs in PB prior to treatment differ between the CG and PG groups and that the number of $Lin^-/CD45^-/CD133^+$ VSELs in PB also differed between the SG and NG subgroups prior to treatment [6]. However, no changes in the number of VSELs circulating in PB were observed in acute anxiety syndrome patients. In parallel, we also measured the plasma levels of factors that modulate the trafficking of these cells, including activated complement cascade components (C3a, C5a, and C5b-9), as well as S1P and SDF-1. Using logistic regression analysis, we found that C3a and S1P are the best predictors of risk and are potential markers for the first psychotic episode [6]. Furthermore, in the SG subgroup, the number of circulating $Lin^-/CD45^-/CD34^+$ VSELs and the S1P plasma level are the best predictors of risk, and we proposed these parameters as novel markers for the first schizophrenic episode [6].

Based on these findings, we propose that altered levels of circulating VSELs, S1P, and C3a in the PB of patients following the first episode of psychosis reveal an accompanying systemic reaction associated with stem cell mobilization and activation of regeneration processes. This latter thesis is supported by increased expression of mRNA for genes characteristic for early neural stem cells detected in the mononuclear cell fraction isolated from PB. Furthermore, we also found that the neuroleptic treatment of the first episode of psychosis does not enhance mobilization of VSELs. Thus, VSELs, S1P, and C3a could potentially be employed as novel markers of the first episode of psychosis [6].

Potential implications of circulating VSELs for psychiatric disorders.

We are aware that further studies are needed to address the role of circulating VSELs in acute psychotic patients. These cells may reportedly differentiate into cells from different germ layers, including neural cells, and thus may play a role in regeneration of brain tissue. We are aware, however, that the potential remodelling of brain tissue during schizophrenia requires further study. In fact, abnormal neurogenesis has been proposed as the cause of certain mental disorders [35]; however, it is unclear whether this occurs during embryonic neurogenesis or in the adult brain.

Further work is also needed to better understand the involvement of factors that modulate the trafficking of stem cells. The results concerning the SDF-1 plasma level in the first episode of psychosis are so far inconclusive, and in fact we did not observe any significant changes in the level of this chemokine in PB [36]. Studies reported in the literature on the role of complement cascade activation are also so far inconclusive [37, 38]. It has been proposed, for example, that complement plays a dual role in schizophrenia: it is neuroprotective in aetiology and neurodegenerative in pathogenesis. Abundant or chronic activation of the CC could, on the one hand, mediate cell damage and lead to neurodegenerative disorders. However, recent studies have implied that C3a is engaged in basal- and ischemia-induced neurogenesis as well as synapse remodelling and pruning [39]. As reported, bioactive C3a and C5a components

of the complement cascade affect neurogenesis, glial chemotaxis, and glial phagocytic activity [35]. Furthermore, the terminal product of complement cascade activation, C5b-C9, also known as the membrane attack complex (MAC), on the one hand, mediates neuronal lysis-related necrosis, and on the other hand, stimulates the proliferation of Schwann cells and inhibits the apoptosis of oligodendrocytes [35-39]. Recently, we demonstrated that activation of the terminal complement cascade and the release of MAC are required for the mobilization of stem cells from BM into PB [40].

The fact that BM-derived cells may also regulate anxiety-like behaviour was recently demonstrated in mice [41]. Specifically, in response to repeated social defeat (RSD) caused by an aggressive intruder, normal mice exhibited anxiety-like behaviour corresponding with an exposure-dependent increase in circulating monocytes as well as cytokine and chemokine responses involved with myeloid cell recruitment and increased presence of monocytes within the perivascular space, parenchyma of the prefrontal cortex, amygdala, and hippocampus. Interestingly, mice deficient in chemokine receptors associated with monocyte trafficking, chemokine receptor 2 (CCR2) knockout or fractalkine receptor (CX3CR1) knockout mice, failed to recruit macrophages to the brain and did not develop anxiety-like behaviour following RSD [41]. This observation provides hints for potential treatment strategies.

Effect of circadian rhythms on circulation of stem cells in PB

It has been demonstrated in mice that enhanced tonus of the vegetative nervous system regulates circadian changes in the mobilization of HSCs into PB. Specifically, circulation of these cells under normal steady-state conditions is regulated by a circadian rhythm, with the peak of these circulating cells occurring in the early morning hours and the nadir at night [25]. In mice exposed to daylight changes, this oscillation in HSCs levels in PB was affected by changes in tonus of the vegetative nervous system [25]. Moreover, in a recent study, it has also been proposed that G-CSF increases sympathetic tonus directly via G-CSF receptors that are expressed on peripheral sympathetic neurons, which would reduce NE reuptake and increase NE availability in the BM microenvironment [28]. Since tonus of the vegetative nervous system may play some role in circadian release of HSCs into PB, it would be interesting to study whether there are any circadian changes in the numbers of these circulating cells in patients suffering from acute psychotic syndromes and acute anxiety syndromes compared with normal controls. Furthermore, a similar analysis of the circadian circulation of HSPCs could be performed in patients medicated with NRI and β2-blockers.

Nevertheless, in one report, no circadian changes in the levels of NE and epinephrine in PB were found that could be explained by a change in circadian rhythms of the patients [42]. NE and epinephrine plasma levels rapidly fluctuated in each normal subject, with no obvious diurnal rhythm. The observed changes were related more to posture and physical activity than to an endogenous circadian surge of catecholamine level regulated by an internal clock [42].

However, while considering the circadian circulation of HSCs, one should also remember that the level of these cells in PB could be affected not only by circa-

dian changes in vegetative nervous system tonus but also by changes in activation of the complement and coagulation cascades. These two important evolutionarily conserved cascades follow circadian changes due to a decrease in blood pH during deep sleep [43, 44]. In support of this possibility, the complement cascade is an important modulator of HSCs trafficking [7]. Furthermore, there is vigorous crosstalk between the coagulation and complement cascades, which are usually simultaneously activated [13]. In particular, thrombin, the final product of coagulation cascade activation, is a potent activator of the C5 component of the complement cascade and, as mentioned above, mobilization of HSCs is severely impaired in C5-deficient mice [26]. Ongoing studies in humans will address which factors modulate circadian release of cells from BM into PB.

Conclusions

Stem cell research and, in particular, the potential cell-trafficking axis between BM and brain requires further study. In this paper, we presented our results suggesting that VSELs play a role in remodelling the brain in schizophrenic patients. Another important question is the potential role of lithium, employed in the treatment of mood instability, on the mobilization of stem cells from BM and their migration to brain, which requires further study [41]. The fact that BM-derived cells may regulate anxiety-like behaviour, as recently demonstrated in normal mice by repeated social defeat caused by an aggressive intruder, lends support to this notion [41]. Therefore, in the coming years we may expect several surprises regarding how circulating stem cells affect pathogenesis and new possibilities for how they might be used in the treatment of psychiatric disorders [45].

References

1. Burns TC, Verfaillie CM, Low WC. *Stem cells for ischemic brain injury: a critical review.* J. Comp. Neurol. 2009; 515: 125-144.
2. Farin A, Liu CY, Langmoen IA, Apuzzo ML. *Biological restoration of central nervous system architecture and function: part 3-stem cell- and cell-based applications and realities in the biological management of central nervous system disorders: traumatic, vascular, and epilepsy disorders.* Neurosurgery 2009; 65: 831-859.
3. Ratajczak MZ, Marycz K, Poniewierska-Baran A, Fiedorowicz K, Zbucka-Kretowska M, Moniuszko M. *Very small embryonic-like stem cells as a novel developmental concept and the hierarchy of the stem cell compartment.* Adv. Med. Sci. 2014; 59: 273-280.
4. Vaccarino FM, Stevens HE, Kocabas A, Palejev D, Szekely A, Grigorenko EL. et al. *Induced pluripotent stem cells: a new tool to confront the challenge of neuropsychiatric disorders.* Neuropharmacology 2011; 60: 1355-1363.
5. Ataka K, Asakawa A, Nagaishi K, Kaimoto K, Sawada A, Hayakawa Y et al. *Bone marrow-derived microglia infiltrate into the paraventricular nucleus of chronic psychological stress-loaded mice.* PLoS One 2013; 8(11); e81744.

6. Kucharska-Mazur J, Tarnowski M, Dołęgowska B, Budkowska M, Pędziwiatr D, Jabłoński M. et al. *Novel evidence for enhanced stem cell trafficking in antipsychotic-naive subjects during their firs tpsychotic episode.* J. Psychiatr. Res. 2014; 49: 18-24.
7. Ratajczak MZ, Kim CH, Wojakowski W, Janowska-Wieczorek A, Kucia M, Ratajczak J. *Innate immunity as orchestrator of stem cell mobilization.* Leukemia 2010; 24: 1667-1675.
8. Ratajczak MZ, Kim CH, Abdel-Latif A, Schneider G, Kucia M, Morris AJ. et al. *A novel perspective on stem cell homing and mobilization: review on bioactive lipids as potent chemoattractants and cationic peptides as underappreciated modulators of responsiveness to SDF-1 gradients*. Leukemia 2012; 26: 63-72.
9. Massberg S, Schaerli P, Knezevic-Maramica I, Kollnberger M, Tubo N, Moseman EA. et al. *Immunosurveillance by hematopoietic progenitor cells trafficking through blood, lymph, and peripheral tissues.* Cell 2007; 131: 994-1008.
10. Wojakowski W, Tendera M, Kucia M, Zuba-Surma E, Paczkowska E, Ciosek J. et al. *Mobilization of bone marrow-derived Oct-4+ SSEA-4+ very small embryonic-like stem cells in patients with acute myocardial infarction.* J. Am. Coll. Cardiol. 2009; 53: 1-9.
11. Kucia M, Zhang YP, Reca R, Wysoczyński M, Machaliński B, Majka M. et al. *Cells enriched in markers of neural tissue-committed stem cells reside in the bone marrow and are mobilized into the peripheral blood following stroke.* Leukemia 2006; 20: 18-28.
12. Paczkowska E, Kucia M, Koziarska D, Halasa M, Safranow K, Masiuk M. et al. *Clinical evidence that very small embryonic-like stem cells are mobilized into peripheral blood in patients after stroke*. Stroke 2009; 40: 1237-1244.
13. Borkowska S, Suszyńska M, Mierzejewska K, Ismail A, Budkowska M, Salata D. et al. *Novel evidence that crosstalk between the complement, coagulation and fibrinolysis proteolytic cascades is involved in mobilization of hematopoietic stem/progenitor cells (HSPCs).* Leukemia 2014; 28(11): 2148-2154.
14. Lapidot T, Kollet O. *The brain-bone-blood triad: traffic lights for stem-cell homing and mobilization.* Hematology Am. Soc. Hematol. Educ. Program 2010; 2010: 1-6.
15. Focosi D, Azzara A, Kast RE, Carulli G, Petrini M. *Lithium and hematology: established and proposed uses.* J. Leukoc. Biol. 2009; 85: 20-28.
16. Levesque JP, Helwani FM, Winkler IG. *The endosteal 'osteoblastic' niche and its role in hematopoietic stem cell homing and mobilization.* Leukemia 2010; 24: 1979-1992.
17. Vandenbosch R, Borgs L, Beukelaers P, Belachew S, Moonen G, Nguyen L. et al. *Adult neurogenesis and the diseased brain.* Curr. Med. Chem. 2009; 16: 652-666.
18. Duan X, Kang E, Liu CY, Ming GL, Song H. *Development of neural stem cell in the adult brain.* Curr. Opin. Neurobiol. 2008; 18: 108-115.
19. Encinas JM, Enikolopov G. *Identifying and quantitating neural stem and progenitor cells in the adult brain.* Methods Cell Biol. 2008; 85: 243-272.
20. Komitova M, Mattsson B, Johansson BB, Eriksson PS. *Enriched environment increases neural stem/progenitor cellproliferation and neurogenesis in the subventricular zone of stroke-lesioned adult rats*. Stroke 2005; 36: 1278-1282.
21. Zhang RL, Zhang ZG, Zhang L, Chopp M. *Proliferation and differentiation of progenitor cells in the cortex and the subventricular zone in the adult rat after focal cerebral ischemia.* Neuroscience 2001; 105: 33-41.
22. Ratajczak MZ, Lee H, Wysoczyński M, Wan W, Marlicz W, Laughlin MJ. et al. *Novel insight into stem cell mobilization-plasma sphingosine-1-phosphate is a major chemoattractant that directs the egress of hematopoietic stem progenitor cells from the bone marrow and its level*

in peripheral blood increases during mobilization due to activation of complement cascade/membrane attack complex. Leukemia 2010; 24: 976-985.

23. Golan K, Vagima Y, Ludin A, Itkin T, Cohen-Gur S, Kalinkovich A. et al. *S1Ppromotes murine progenitor cell egress and mobilization via S1P1-mediated ROS signaling and SDF-1 release*. Blood 2012; 119: 2478-2488.
24. Martin R, Sospedra M. *Sphingosine-1 phosphate and central nervous system.* Curr. Top. Microbiol. Immunol. 2014; 378: 149-170.
25. Katayama Y, Battista M, Kao WM, Hidalgo A, Peired AJ, Thomas SA. et al. *Signals from the sympathetic nervous system regulate hematopoietic stem cell egress from bone marrow*. Cell 2006; 124: 407-421.
26. Lee HM, Wu W, Wysoczyński M, Liu R, Zuba-Surma EK, Kucia M. et al. *Impaired mobilization of hematopoietic stem/progenitor cells in C5-deficient mice supports the pivotal involvement of innate immunity in this process and reveals novel promobilization effects of granulocytes.* Leukemia 2009; 23: 2052-2062.
27. Wysoczyński M, Ratajczak J, Pędziwiatr D, Rokosh G, Bolli R, Ratajczak MZ. *Identification of Heme Oxygenase 1 (HO-1) as a Novel Negative Regulator of Mobilization of Hematopoietic Stem/Progenitor Cells.* Stem Cell Rev. 2014; DOI 10.1007/s12015-014-9547-7.
28. Lucas D, Bruns I, Battista M, Mendez-Ferrer S, Magnon C, Kunisaki Y. et al.. *Norepinephrine reuptake inhibition promotes mobilization in mice: potential impact to rescue low stem cell yields.* Blood 2012; 119: 3962-3965.
29. Bonig H, Papayannopoulou T. *Hematopoietic stem cell mobilization: updated conceptual renditions.* Leukemia 2013; 27: 24-31.
30. Albus M, Ackenheil M, Engel RR, Muller F. *Situational reactivity of autonomic functions in schizophrenic patients.* Psychiatry Res. 1982; 6: 361-370.
31. Cai HL, Fang PF, Li HD, Zhang XH, Hu L, Yang W. et al. *Abnormal plasma monoamine metabolism in schizophrenia and its correlation with clinical responses to risperidone treatment.* Psychiatry Res. 2011; 188: 197-202.
32. Sheehan DV, Lecrubier Y, Sheehan KH, Amorim P, Janavs J, Weiller E. et al. *The Mini-International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview for DSM-IV and ICD-10.* J. Clin. Psychiatry 1998; 59(supl. 20): 22-33, quiz 34-57.
33. Rzewuska M. *Validity and reliability of the Polish version of the Positive and Negative Syndrome Scale (PANSS).* Int. J. Methods Psychiatr. Res. 2002; 11: 27-32.
34. Kucharska-Mazur J, Pędziwiatr D, Poniewierska A, Tkacz M, Suszyńska M, Tarnowski M. et al. *A lack of positive effect of enhanced vegetative nervous system tonus on mobilization of hematopoietic stem and progenitor cells in patients suffering from acute psychotic syndromes.* Leukemia 2013; 27: 959-961.
35. Buffo A, Rolando C, Ceruti S. *Astrocytes in the damaged brain: molecular and cellular insights into their reactive response and healingpotential.* Biochem. Pharmacol. 2010; 79: 77-89.
36. Fernandez-Egea E, Bruna A, Garcia-Rizo C, Bernardo M, Kirkpatrick B. *Stem cell signaling in newly diagnosed, antipsychotic-naïve subjects with nonaffective psychosis.* Mol. Psychiatry 2009; 14: 989-991.
37. Boyajyan A, Khoyetsyan A, Chavushyan A. *Alternative complement pathway in schizophrenia.* Neurochem. Res. 2010; 35: 894-898.
38. Hakobyan S, Boyajyan A, Sim RB. *Classical pathway complement activity in schizophrenia.* Neurosci. Lett. 2005; 374: 35-37.

39. Stevens B, Allen NJ, Vazquez LE, Howell GR, Christopherson KS, Nouri N. et al. *The classical complement cascade mediates CNS synapse elimination.* Cell 2007; 131: 1164-1178.
40. Kim CH, Wu W, Wysoczyński M, Abdel-Latif A, Sunkara M, Morris A. et al. *Conditioning for hematopoietic transplantation activates the complement cascade and induces a proteolytic environment in bone marrow: a novel role for bioactive lipids and soluble C5b-C9 as homing factors.* Leukemia 2012; 26: 106-116.
41. Wohleb ES, Powell ND, Godbout JP, Sheridan JF. *Stress-induced recruitment ofbone marrow-derivedmonocytes to the brainpromotesanxiety-like behavior.* J. Neurosci. 2013; 33: 13820-13833.
42. Schofl C, Becker C, Prank K, von zur Muhlen A, Brabant G. *Twenty-four-hour rhythms of plasma catecholamines and their relation to cardiovascular parameters in healthy young men.* Eur. J. Endocrinol. 1997; 137: 675-683.
43. Reis ES, Lange T, Kohl G, Herrmann A, Tschulakow AV, Naujoks J. et al. *Sleep and circadian rhythm regulate circulating complement factors and immunoregulatory properties of C5a.* Brain Behav. Immun. 2011; 25: 1416-1426.
44. Wolk R, Gami AS, Garcia-Touchard A, Somers VK. *Sleep and cardiovascular disease.* Curr. Probl. Cardiol. 2005; 30: 625-662.
45. Ratajczak MZ, Jadczyk T, Pędziwiatr D, Wojakowski W. *New advances in stem cell research: practical implications for regenerative medicine.* Pol. Arch. Med. Wewn. 2014; 124: 417-426.

Address: Mariusz Z. Ratajczak
Stem Cell Institute, James Graham Brown Cancer Center University of Louisville 500 S. Floyd Street, Rm. 107 Louisville, KY 40202, USA