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Chapter

Alternative Immune-Mediated-Based Methods in the Aplastic Anemia Treatment

Vivian Gonzaga, Bruna Policiquio, Cristiane Wenceslau and Irina Kerkis

Abstract

Acquired aplastic anemia (AA) is characterized by partial or total bone marrow (BM) destruction resulting in pancytopenia. Most of the acquired AA is the result of autoimmune condition the imbalance between T-regulatory cells (Treg), abnormal cytokines production and cytotoxic T cells activation, leading to the hematopoietic stem cells (HSCs) death. The first-line treatment is given by HSC transplant, but some patients did not respond to the treatment. Therefore, new technologies need to treat AA nonresponder patients. Studies are in progress to test the efficacy of stem cell-based therapeutic as mesenchymal stem cells (MSCs), which confer low immunogenicity and are reliable allogeneic transplants in refractory severe AA cases. Furthermore, MSCs comprise the BM stromal niche and have an important role in supporting hematopoiesis by secreting regulatory cytokines, providing stimulus to natural BM microenvironment. In addition, MSCs have immunomodulatory property and are candidates for efficient supporting AA therapy.

Keywords: allogeneic transplant, mesenchymal stem cell, immune-mediated aplastic anemia, paracrine effects, immunomodulation

1. Introduction

Aplastic anemia (AA) is a rare disease, caused by bone marrow (BM) aggression resulting in hypo or aplastic BM with precocious fat replacement and consequently to peripheral blood pancytopenia [1, 2]. The autoimmunity process in AA occurs due to the activation of the oligoclonal cytotoxic T cells that will lead the hematopoietic cells to apoptosis. Its triggering occurs by the imbalance between CD8 +, CD4 +, T-Helper (Th), Th type 1 (Th1), Th type 2 (Th2), Th17 type (Th17), Natural Killer (NK) and T-regulatory cells (Treg). Besides, there is also an abnormal production of proinflammatory cytokines, such as tumor necrosis factor-alpha (TNF-α), interferon-gamma (IFN-γ) and transformed growth factor (TGF) [3–7].

For severe cases, immunosuppressive therapy is accepted as the first-line treatment option and the allogeneic transplantation of BM and hematopoietic stem cells (HSCs). However, 30–40% of patients with severe aplastic anemia (SAA) remain pancytopenia following the treatment. The transplant option still has a restricted number of compatibility between suitable donors. Additionally, patients aged >50 years are not eligible for transplant [8].

A new viable alternative for the treatment of AA has been sought and the use of mesenchymal stem (MSCs) therapy may be a promising therapeutic candidate mainly because of their hypoimmunogenicity and the lack of rejection after transplants and immunomodulatory effects, which may promote decreasing the symptoms of the disease [9, 10]. These benefits are attributed to the paracrine effects, above all by its ability to regulate the immune system [11].

Actually, is known that MSCs have wide therapeutically potential attributed by paracrine effects and the past decades have seen explosion research directed to understand better these MSCs mechanism and function [12]. One of the main and most important features of MSCs is the low expression of human leukocyte antigen (HLA) class I, with no expression of HLA class II. This feature allows the cell to be characterized as hypoimmunogenic, since it does not stimulate the patient's immune system and can be used safely in transplants [13]. More recently, the studies showed that the main cause of AA is autoimmunity. Through the secretion of bioactive molecules, MSCs have the capacity of regulating immune responses. The mechanism of MSCs may decrease secretion of proinflammatory cytokines such as transforming growth factor (TGF), IFN-γ TNF-α, interleukin (IL)-17 and increase secretion of many soluble mediators, including anti-inflammatory cytokines stimulation that inhibit antigen-presenting cells (APCs) functions, which are capable to decrease proliferation of dendritic cells (DCs) and regulate macrophage activity by polarizing proinflammatory phenotype (M1) to anti-inflammatory phenotype (M2) [14, 15]. Therefore, the decrease of B cells proliferation and antibodies production and adjustment of T cells activities as well as inhibit the proliferation of cytotoxic T cells and stimulate Treg activity [16].

MSCs therapy has gained space due to its vast therapeutic potentials such as immunomodulation mechanisms and main safety as bioproduct. Thus, this chapter will discuss the challenges of allogeneic MSCs as an alternative for an efficient therapeutic in AA immune-mediated treatment.

2. Aplastic anemia: general features

AA is a disorder characterized by BM hypocellularity, and peripheral blood pancytopenia due to a deficit of HSCs. It affects mostly children, young adults, and adults, over 60 years of age [17]. This condition can be similar to other hematologic disorders, however, in most cases, the AA is caused by reduced HSCs function, an increase in HSCs apoptosis level, consequently, the decreased of HSCs and hematopoietic progenitors and lastly, microenvironment fat replacement [18, 19].

Following the patient diagnosis, AA can be considered as moderate or severe. The patients with pancytopenia may present symptoms of anemia purpura or skin hemorrhage, and in most of the cases there is an infection association, that may worsen the symptoms [20]. Three main criteria are used for the diagnostic: neutrophil count lower than 0.5×10^9 cells/L, reticulocyte count lower than 1% and platelet count lower than 20×10^9 cells/L [21]. To confirm acquired AA, the clinical case must be differentiated from other hematological diseases, as well as from the signs of malignant cell transformation or myelodysplasia [22].

Normally the first AA etiology is uncertain and for this reason, the disorder is considered heterogeneous in origin and characterized as idiopathic [23]. AA is associated with exposures to chemical agents (pesticides and benzene), cytotoxic drugs (antineoplastics, antibiotics, non-steroidal anti-inflammatory drugs), active viral infections exposure (Epstein Barr, hepatitis virus, human immunodeficiency virus parvovirus) and radiation exposure [18, 24, 25]. However, these causes considered

secondary etiologies, since the studies are directed to the primary etiology of AA to autoimmunity [26, 27]. AA pathogenesis involves an immunity dysfunction, initially provoked by the activated T cells [23], which leads to an abnormal hematopoietic microenvironment, destruction of hematopoietic stem/progenitor cell and differentiation deficiency. These findings suggest that the immune system plays an important role in the pathogenesis of AA.

2.1 AA pathophysiology

Currently, the studies of AA etiology are focused on the immune mechanism of hematopoietic cells destruction. Many researchers [28, 29–31] have demonstrated that the dysfunction of T cells might be a key factor in recent characterization as an autoimmune disease [28]. Most of the acquired AA is the result of an immune-mediated process as an imbalance between CD8+ and CD4+ T cells, including Th1, Th2, Treg and Th17 cells, NK, and natural killer T cells (NK T) that leads to apoptosis of BM cells triggered by cytotoxic T cells activation [6, 17].

The abnormal immunoregulatory cell functions observed in AA can be attributable to abnormal antigen stimulation and some inappropriate T cells activation [28]. Studies demonstrated that patients with AA have a significantly increased proportion of Th1 cells, and showed a reduced fraction of natural killer T cells and regulatory T cells, together with an increased level of TNF-α, a consequent elevation of IL-6, IL-8, and IL17 productions [18]. Additionally, there is also an abnormal production of proinflammatory cytokines including IFN-γ and TGF [4, 5, 28, 32]. The new T cells subset was characterized as Th17 and currently is known that both Th17 cells and the cytokine IL-17, which is secreted by Th17 cells, also is in an association with AA pathogenesis [31]. Studies showed that AA patients who presented an increase in the frequency of Th17 cells had a positive correlation with an increase in the IFN- γ and IL-17 expression. Autoimmunity promotes inflammatory Th17 immune responses that contributed to disease pathophysiology [29].

Otherwise, AA is attributed to inappropriate antigen stimulation and abnormal APCs activation [28], resulting in the priming of T cells specific for hematopoietic cells [33, 34]. APCs exhibit a significant increase in the expression of major histocompatibility class 2 (MHCII), increasing the recognition of CD4+ T cells. In AA, T cells are also stimulated by unknown antigens or abnormal APC activation as DCs and macrophages, which trigger a series of immune responses. Studies have shown that immunoregulatory cell dysfunction leads to a corresponding immune tolerance disorder and renders the body unable to recognize autologous hematopoietic cells [28].

Although the definitive mechanism has not been identified, some genetic factors are the targets of ongoing research, such as the molecular basis of the aberrant immune response and hematopoietic cell deficiency, telomere repair gene mutations in the target cells and unregulated T cell activation pathways and cytokine genes polymorphisms [9, 26, 28]. These changes in the nucleotide sequence and gene regulation are associated with an increased immune response and suggest a genetic basis for aberrant T cells activation in BM failure [35].

2.2 AA treatment

The treatment depends on the severity of the disease, once for moderate cases are based on red blood cell (RBC) transfusions, on platelet transfusions to prevent bleeding, and on supportive care in association with antibiotic aiming to reestablish blood cell volume and prevent secondary infections [17]. However, the pancytopenia of many moderate cases may progress to severe [21]. For severe cases,

immunosuppressive therapy is accepted as a first-line treatment option. However, 30–40% of patients with SAA remain pancytopenia following the treatment. Patients with SAA, which are refractory or have a relapse after immunosuppressive treatment, may undergo allogeneic hematopoietic stem cells transplantation (HSCT). However, about one-third of patients do not have a suitable donor for HSCT. Additionally, patients aged >50 years are not eligible for transplant [8].

Furthermore, the immunosuppressive drug treatment has several side effects on patients. On the other hand, the patients often do not respond adequately to the therapies and are not suitable for life treatment (refractory patients) [24]. Therefore, immunosuppressive drugs are considered supporting AA treatment, once it does not promote the cure [20].

2.3 Allogeneic transplantation and alternative methods for AA treatment

Generally, patients are treated with allogeneic HSCs or whole BM transplantations, which replace since HSCs, hematopoietic precursors, until differentiated bloodstream cells and immune system cells. However, in all types of transplants, the treatment involves a combination of immunosuppressive agents or radiation therapy to prevent and to eliminate residual host BM [24]. The transplantation success varies according to risk factors, such as age and mainly histocompatibility allogeneic HLA-matched sibling donors, which are rare for the majority of patients. Despite being well established for many years, the transplanted patients can trigger late complications, such as the development of graft versus host disease (GVHD) and infections, especially in patients who have received hematopoietic grafts from HLA antigen matched donor [36, 37]. Studies show that the incidence of GVHD after unrelated donor transplantation can achieve ~14%, and overall survival index was 57% for all 8 HLA-loci matched transplants and 39% for 1-loci mismatched transplant [38]. Thus, for BM and HSCT, the immediate challenge is the extension of stem cell therapies to all patients, regardless of age, with a histocompatible sibling [24].

Since then a new viable alternative for the treatment of AA has been sought and the use of MSCs transplantation becomes of choice. The MSCs therapy may be a promising therapeutic candidate mainly because of their hypoimmunogenicity, the lack of rejection after transplants and immunomodulatory effects, which may promote decreasing the symptoms of the disease [39]. These benefits are attributed to MSCs paracrine effects, above all to their ability to regulate the immune system. MSCs may help for AA treatment, especially for autoimmune type [11].

3. Mesenchymal stem cell: general features

MSCs are multipotent progenitors, which were first isolated from an adult organism by Friedenstein and colleagues in 1968, and described years later by Caplan and colleagues [40, 41]. These cells include firstly an inherent autocrine effect, as self-renewal and differentiation potential for a variety of cell types, as main adipocytes, osteoclasts, and chondrocytes [42], depending on the surrounding microenvironment conditions [43]. Currently, such cells have shown to be isolated from many postnatal and adult tissues, such as adipose tissue, umbilical cord, placenta, dental pulp, and others [44, 45].

Initially, the mechanism therapeutic potential of the MSCs was based only on the potential for regeneration through cellular self-renewal and its plasticity.

Further studies have shown low engraft of MSCs in injured areas that questioned the hypothesis that MSCs repair tissue damage by replacing cell loss with newly differentiated cells [46, 47].

3.1 MSC: paracrine effects

It is known that MSCs have wide therapeutically potential attributed to paracrine effects and the past decades explosion research was directed to understand better these MSCs mechanism and function [12]. Although the therapeutic mechanisms of MSCs are not yet well characterized, it is possible to say that their paracrine effects consist in the secretion of bioactive molecules such as a variety of cytokines and growth factors as like anti-inflammatory, anti-apoptotic and angiogenic [46–51].

MSCs can to migrate to the lesion site through signals from specific chemokines. This process called homing consists of the steps of activating adhesion molecules, rolling to the endothelium, adhesion, and migration to the tissue that is the source of chemokine inflammation production [52, 53]. The current hypothesis is that paracrine factors secreted by MSCs promote protective microenvironment and repair by local tissue-resident progenitor populations, favoring the hypothesis of detecting favorable effects even in the absence of the cells at lesion sites [54].

3.2 MSC: immunogenic effect and safety for transplantation

One of the main and most important features of MSC is the low expression of HLA class I, with no expression of HLA class II. Also, MSCs do not appear to express the co-stimulatory molecules CD80 or CD86 required for effector T cell induction [55]. The absence of co-stimulatory molecules implies that any residual engagement of the T cell receptor on Th cells would result in absence of the normal immune response to a particular antigen and contribute to tolerance rather than allogeneic responses. This feature allows the cell to be characterized as hypoimmunogenic, since it does not stimulate the patient's immune system and can be used safely in transplants [113. As well, MSCs have properties attributed to immune functions, indicating their ability to immunomodulatory activity. Studies indicated that MSCs can regulate immune responses during chronic inflammation through the innate and adaptive immune system, regulating the recruitment and their function [56, 57].

3.3 MSC: immunomodulatory potential

The paracrine effects of MSCs may have great importance in the treatment of autoimmune diseases. Through the secretion of bioactive molecules, MSCs have the capacity of regulating immune responses. These cells can regulate adaptive immune responses through multiple redundant pathways, interacting with various immune cells and secreting soluble mediators such as IL-6, IL-10, prostaglandin E2 (PGE2), nitric oxide (NO), transforming growth factor-β1 (TGF-β1), and hepatocyte growth factor (HGF), indoleamine-pyrrole 2, 3-dioxygenase (IDO) [58, 59]. They can regulate APCs activity, decreasing maturation and proliferation of DCs [14]. MSC also may regulate macrophage activity by polarizing its pro-inflammatory phenotype (M1) to its anti-inflammatory phenotype (M2) [15]. Therefore, suppress T cell proliferation and activation and regulate the differentiation of Th cells and act on the humoral response by inhibiting of B cell activation and antibody production [60]. MSCs may also reduce pro-inflammatory cytokines proliferation, such TNF-α, which has an important role of the pathogenesis of autoimmune diseases and chronic inflammation (**Figure 1)** [14, 16, 61].

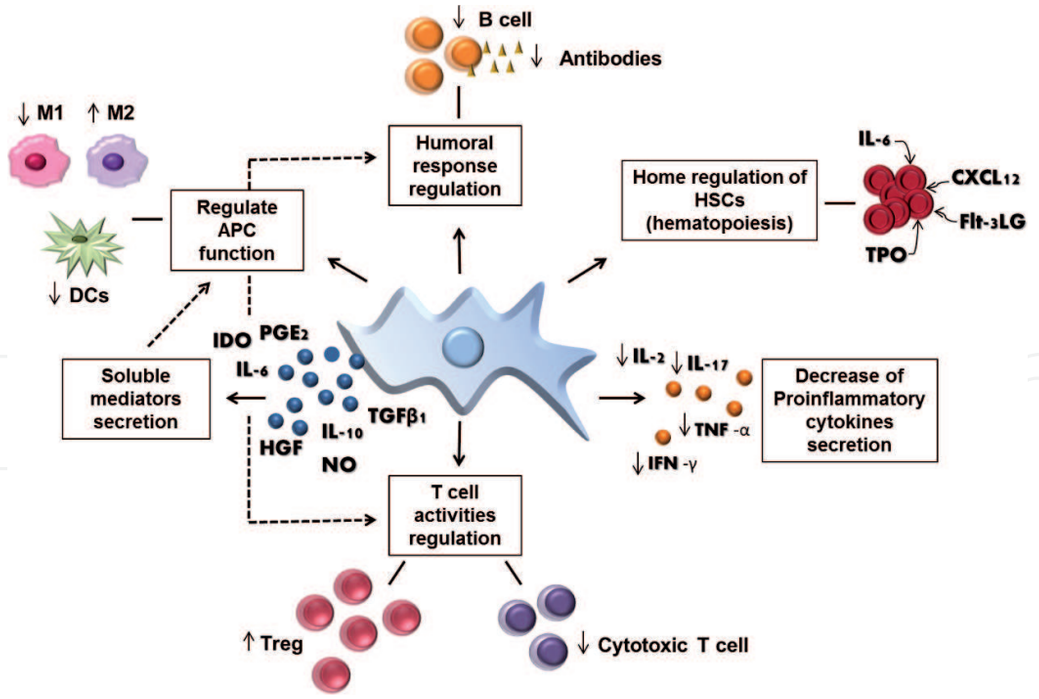

Figure 1.
Benefits of MSCs paracrine effect (immunomodulatory) on immune cells imbalance. MSCs secrete many soluble mediators, including anti-inflammatory cytokines stimulation that regulates APCs functions capable to decrease proliferation of DCs and regulate macrophage activity by polarizing proinflammatory phenotype (M1) to anti-inflammatory phenotype (M2). Therefore, they are responsible for humoral response regulation by the decrease of B cells proliferation and antibodies production. The APCs are also capable to regulate the T cell activities as well as inhibit cytotoxic T cell proliferation and upregulation and increase of Treg cells. MSCs may also promote the decrease of proinflammatory cytokines secretion. And act on the homing regulation of HSCs mechanism on stages of adhesion, expansion, and migration through chemokine and other factors secretion.

4. MSC mechanism in AA treatment

The first paracrine effect, showed for MSCs, was the capacity to support HSCs growth *in vitro*. Afterward, adipose tissue (AT) – derived MSCs also supported HSCs growth *in vitro* [62, 63]. Therefore, the most successful clinical application of MSCs is involved in the hematological disease.

At BM microenvironment, MSCs niche supports hematopoietic cells and produce factors recruiting HSCs and supporting hematopoiesis [64]. This mechanism occurs through chemokine secretion of C-X-C motif chemokine ligand 12 (CXCL12), which acts on the homing regulation of HSCs, regulating the stages of adhesion, expansion and migration [65, 66]. The secretion of other factors is also important in the proliferation of HSCs mechanisms such as Flt-3 ligand (FLT3LG) [67], thrombopoietin (TPO) [[68] and IL-6 [17]. That despite being a proinflammatory cytokine in general, when IL-6 is secreted in BM microenvironment, is capable to stimulate hematopoiesis [69, 70].

More recently, the studies showed that the main cause of AA is autoimmunity. This process occurs in the result of an imbalance between CD8 + and CD4 + T cells, including Th1, Th2, Th17, NK, leading to the death of hematopoietic cells and their precursors [28]. Many studies have hypothesized that the onset of the immune imbalance in AA begins by stimulating APCs through an unknown antigen resulting in the T cells activation [71]. Another important mechanism of MSCs is the immunomodulation mechanism. MSCs can act directly on AA imbalance by T cells suppression, inhibiting activation and proliferation of T cells [72]. MSCs also inhibit the secretion of two important cytokines present in the pathology of AA, the

AA disorders x MSC benefits	
Aberrant secretion of pro-inflammatory cytokines ↑IFN-γ ↑TNF-α ↑IL-17 ↑IL-2;	Immunomodulatory effect: Decreased secretion of proinflammatory cytokines, ↓IFN-γ ↓TNF-α ↓IL-17 ↓IL-2;
Imbalance between CD8+ and CD4+ T cells; ↑ Cytotoxic T cell ↓Treg	Regulation of T cell activity and Treg cell proliferation ↓ Cytotoxic T cell ↑Treg
Apoptosis of HSC and progenitor cells ↑ Apoptosis	Protect BM by antiapoptotic properties ↓ Apoptosis
BM hypoplasia	Recovery of BM ↑ Hematopoiesis improvements ↑ CXCL12 ↑FLT3LG ↑TPO ↑IL-6
Abnormal APC activation ↑ DCs ↑ Macrophage	Regulate APCs functions ↓ DCs maturation and proliferation and ↓ Macrophages M1 activation ↑ Macrophages M2 activation
Abnormal humoral response ↑ B cells ↑ Antibodies production	Regulate humoral response ↓ B cells ↓ Antibodies production
Irregular activity of NK cells ↑ Cytotoxicity of NK cells	↓ Cytotoxicity of NK cells

MSC can decrease secretion of pro-inflammatory cytokines such as TGF, IFN-γ TNF-α, IL-17, regulate T cell activity, inhibit proliferation of cytotoxic T cells and stimulate Treg activity. MSC has anti-apoptotic properties, protects BM environment and recovery BM through cytoprotective effect and stimulates macrophages M2 activation and hematopoiesis improvements. MSCs may also regulate APCs functions, humoral response, and cytotoxicity of NK cells.

Table 1.
Table shows disorders characterized in AA and the mechanism of action of MSCs in AA pathology.

INF-γ and TNF-α and stimulate the proliferation of Treg, promoting the production of the anti-inflammatory cytokine IL-10 **Table 1** [73, 74]. In addition, some studies also show that MSCs also acts through its anti-apoptotic effects [75].

5. Biodistribution and engraftment of allogeneic MSC in BM

In the last years, several studies have been exploring intravenous administrations (IV) due to being safe and do not present morbidity risk for patients. However, still lack the data about the biodistribution mechanism of MSCs and about how these cells engraftment on the target organ, which is essential for the success of clinical studies. It is known that the biodistribution is influenced in vivo and in vitro conditions. Stromal cell-derived factor 1 (SDF-1) (also known as CXCL12) is upregulated at sites of injury and acts as a chemoattractant to recruit circulating or residing MSCs expressing its cognate receptor CXC chemokine receptor 4 (CXCR4). It has been demonstrated that the CXCR4-SDF-1 axis is critical for BM homing [76]. Diverse studies demonstrate that some in vitro conditions may influence the expression of adhesion molecules [77, 78]. For instance, long expansion periods [79] and cells culturing at high density may reduce CXCR4 cell expression; the cells cultured at higher confluence secrete more metalloproteinase inhibitor 3, which decreases migration of MSCs when compared to those cultured at the low confluence [80]. Hypoxia condition may increase CXCR4 expression; on the other hand, hypoxia may decrease matrix metalloproteinase-2 secretion and an increase in membrane-type 1 matrix metalloproteinase [81].

In vivo engraftment is influenced by interactions of MSCs with different types of immune cells that depend on their ability to respond to signals from the immune

system. On the other hands, the MSCs biodistribution and homing depend on the host niche. Interesting the MSCs migration and homing to target tissue can be influenced positively by irradiation. It has been demonstrated an increased absolute number of human MSCs in the brain, heart, bone marrow, and muscles after total body irradiation and MSCs IV administrations in mice, when compared to the untreated control [82].

Some animal studies evidence that MSCs can engraftment in BM after systemic administration [83]. Studies in patients showed MSCs engraftment into BM 30 days after the second MSCs IV administration. Although, after MSCs infusion was observed no recovery of hematopoietic tissue, interstitial hemorrhage, edema, and all adipocytic necrosis disappeared in BM [84]. Other studies indicate the engraftment due to myeloid and plated recovery after HSCs and MSCs transplantation [85, 86].

6. MSC use in clinical studies

6.1 Clinical potential and market of MSC in hematopoietic disorders

MSCs have been implicated in immunomodulatory therapy, in particular, in GVHD treatment and as an adjunct to hematopoietic stem cell transplantation (HSCT) to help enhance engraftment [87, 88]. The first major clinical trial of MSCs (Prochymal) was for the treatment of steroid-refractory of GVHD (NCT00366145) [89, 90]. The primary endpoint of the study was complete remission at day 28 after allogeneic BM-MSCs infusion but was not significantly increased compared to placebo [89, 91]. In 2012, MSCs have bens conditional approval to treat children GVHD in Canada, based on subset analysis that suggested children with GVHD were responsive to MSCs [89, 92, 93]. Many new studies have been developed in recent years; however, a few of them have attempted to look at biological correlates of response to therapy. Isolated studies reported serum biomarkers of GVHD severity including IL-2, *tumor necrosis factor receptor 1 (*TNFR1), regenerating islet-derived protein 3 alpha (Reg3a), and levels of inflammatory cytokines, which not clearly correlate with the response in humans. More studies are needed to obtain correlative research data [94, 95]. This outcome results in the first Food and Drug Administration (FDA) approved MSCs product in the United States [96, 97].

6.2 Clinical studies with MSC in combination to HSCT transplantation for treat AA

Cotransplantation of HSCs with (umbilical cord) UC-MSCs has been performed to study whether the last will be able to support hematopoiesis, enhance the engraftment of HSCs, and reduce the incidence of GVHD following HSCT [98–100]. These studies include adult and children in AA patients [101, 102]. Stem cells application was mainly intravenous. In some of the studies multiple (five) infusions were used. All clinical protocols have been developed in the presence of traditional immunosuppressive protocol to prevent GVHD manifestation [98–102].

One pioneer study, where the conditioning of patients was myeloablative or reduced, followed BM-MSCs treatment together with allogeneic HSCT. This study showed that co-transplantation of MSCs resulted in fast engraftment of absolute neutrophil count and platelets and 100% donor chimerism [87]. In turn, Yamei and co-workers (2014) demonstrate prolonged survival (follow up of 78 months) in 80.9% patients after cotransplantation of the culture-expanded third party donor-derived UC-MSCs in 21 young people with SAA undergoing haplo-HSCT [103]. Even so, the patients did not show infusion toxicity. This study showed that MSCs support *in vivo*

normal hematopoiesis and display potent immunosuppressive effects. The other metacentric study shows that cotransplantation of BM-MSCs and haplo-HSCT could reduce the risk of graft failure and severe GVHD in SAA [104]. Similar data were obtained in a study that used cotransplantation of haploidentical HSCs and BM-MSCs into children with SAA without an HLA-identical sibling donor. It was shown that such cotransplantation seems to be safe and may improve survival rates and reduce the risk of graft failure [105]. Another multicenter study, which explored cotransplantation of BM-MSCs with allo-HSCT, reported that such treatment could ameliorate clinical outcomes of a GVHD, viremia, and survival in allo-HSCT for AA patients [106].

6.3 Clinical studies with MSC for treat AA

Nowadays there a few clinical studies using only MSCs single to treat AA. All studies used MSCs isolated from BM s and adult patients with severe or non-severe AA and refractory. The via of MSCs administration used was IV and the number of administrations was 2 until 5 depending on study in combination with conventional immunosuppressive therapy.

The study development by Pang et al. showed, six of 18 patients (33.3%) achieved a complete response or a partial response to MSCs treatment [107]. In six patients, two achieved a complete response including recovery of three hematopoietic cell lines after MSCs therapy. Similar results was achieved by Cle et al. 2015 using MSCs being 22% of all patients (18 patients) presents hematologic response at 6 months after MSCs transplantation [108]. One clinical trial phase II conducted in China evaluated the MSC overall response rate and safety using a significant number of refractory AA younger patients (n = 72). The study performed full quality control of BM-MSCs production, which includes counts, viability, morphology, endotoxin, aseptic culture, immunophenotype. It was the first clinical study that showed significant results in BM functional recovery. The rate response of patients was 28.4% being that 6.8% complete response and 21.6% partial response after MSCs transplantation. Among patients with hematologic response, ten patients had normalization of cellularity BM followed for more than 1 year MSCs transplantation. Seven patients got adverse events such as fever and headaches. No other adverse events were observed in the study. At the follow-up endpoint, nine patients died. One patient with RAEB-II died of disease progression, two patients died of intracranial hemorrhages, and six patients died of serious infection [107]. In other two studies were reported adverse events such as, fever, hypoxemia, mild dyspnea and diarrhea during MSCs administration or some hours after MSCs injection, this phenomenon occurs in 2 of 16 patients [107] and 7 of the 18 patients [108]. None major adverse effects were reported in all studies during months of follow-up of each respective study. Fuillard et al., 2003 reported one death due to fungal infection and Cle et al. 2015 four patients died in consequence result of heart failure and bacterial or invasive fungal infections and none of the deaths in both studies were directly attributable to MSCs infusions [84, 108].

These preliminary studies support the concept that MSCs replacement can improve BM stroma and may alleviate symptoms severe and non-severe AA patients. However, larger studies with a significant number of patients are needed to evaluate the utility of MSCs further.

7. Conclusion and future perspectives

The progress in dissecting the underlying and complex pathophysiology of AA has been gain space over the past years in the hematology research community [26]. In addition to that, the need for an optimal alternative of a targeted treatment for

this disorder. It is too soon to place the conventional AA treatment methods, but MSCs have gained space for demonstrating positive results in several AA clinical studies and other hematological diseases. The hypoimmunogenicity advantages, ensuring the absence of rejection in patients due to no expression of MHC class II, prevention and treatment of GVHD traditional transplants, and mainly immunomodulatory action presented [109]. Essential in the environment imbalance provoked by own immune system in people committed by the AA disorder. The MSCs are able in a modulating way to relieve the BM self-attack [110].

Contemporary, personalized therapies are famous in the whole scientific world. The MSCs may fit into this class due to their paracrine effects. These cells can assist in diverse situations such as: migration, injury recovery, stimulates cells renewal, death cell prevention, anti-inflammatory and modulation of the immune system to control the autoimmune environment [111]. Thus, MSCs have the heterogeneous capacity in varied therapies field. And the patient may have alternative use according to their needs.

In that event, the current way is providing the MSCs safety and acceptance by regulatory agencies as new biological product [112], which has already been proven to be more efficient than synthetic industries products [113]. And finally, implement the MSCs as ideal allogeneic transplant model, even for adequacy periods used as support for other established therapies.

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Conflict of Interest

The authors declare no conflict of interest.

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