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Immune-Mediated Cytopenias After Hematopoietic Cell Transplantation: Pathophysiology, Clinical Manifestations, Diagnosis, and Treatment Strategies

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Abstract

Purpose of Review Discuss the pathophysiology, clinical presentation, diagnosis, and treatment of immune-mediated cytopenias (IMC) after hematopoietic cell transplantation (HCT).

Recent Findings Key risk factors for post-HCT IMC include younger age, non-malignant disease, and umbilical cord blood stem cell source. While anemia predominates, any or all three hematopoietic cell lines can be affected. In rare cases, IMC can cause graft failure or death. IMC is hypothesized to result from immune dysregulation upon reconstitution of donor hematopoietic cells (i.e., dysfunctional regulatory T cells). Aside from blood product transfusions, IMC treatment includes immune-suppressive or ablative agents. First-line therapies, including corticosteroids and intravenous immunoglobulin, are often inadequate, prompting use of additional agents aimed at antibody production/T cell dysfunction or direct antibody removal via plasmapheresis.

Summary IMC occurs in up to 20% of high-risk HCT populations. Morbidity and mortality from IMC post-HCT have been reduced by improved recognition and aggressive early interventions.

Keywords Hematopoietic cell transplantation · Bone marrow transplantation · Immune-mediated cytopenias · Anemia · Thrombocytopenia · Neutropenia

Introduction

Immune-mediated cytopenias (IMC) are increasingly recognized as an important complication in patients receiving either allogeneic or autologous hematopoietic cell transplantation (HCT). While often referred to as autoimmune cytopenias, the pathophysiology in the post-allogeneic HCT setting is not clearly allo- or auto-immune, hence our preference for the term *immune-mediated cytopenias*. Historically, these immune-mediated conditions were associated with significant morbidity and mortality. However, early diagnosis and prompt management have led to improved outcomes [1, 2, 3•, 4•]. Herein, we review the pathophysiology, risk factors, clinical presentation, diagnosis, and treatment of IMC in the post-transplant setting.

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Cellular Engraftment and Immune Reconstitution Following Hematopoietic Cell Transplantation

Understanding the kinetics of immune, specifically lymphocyte, reconstitution is critical to comprehending the pathophysiology of post-transplant IMC. Neutrophils are the first cells of immunity to recover following autologous transplant, approximately 10 days post-transplant [5]. In patients undergoing allogeneic HCT, neutrophil engraftment is dependent on the source of hematopoietic stem cells (i.e., approximately 14 days following peripheral blood cells, 21 days following bone marrow cells, and 30 days following umbilical cord blood cells) [6, 7].

Early post-HCT, T cell populations are comprised exclusively of residual recipient memory T cells that survived the conditioning regimen and homeostatic or disease-specific proliferating donor T cells [8, 9]. De novo production of engrafted donor lymphocyte populations by T cell progenitors, or neo-thymopoiesis, often takes greater than 100 days after allogeneic HCT. Full lymphocyte recovery, including B cell reconstitution, may take up to 2 years post-HCT [6, 7]. Regulatory T cells (Tregs), which are crucial to reducing inappropriate immune activation and allowing for antigen self-tolerance, may also remain reduced compared to healthy controls up to 2 years after transplantation [7, 10]. Of note, lymphocyte recovery occurs much sooner in autologous transplant patients given reduced in vivo immuno-ablation by serotherapy during conditioning and the lack of immunosuppressive therapy for graft-versus-host disease (GvHD) prophylaxis [8].

Epidemiology

Large retrospective studies of pediatric and adult patients undergoing allogeneic HCT for malignant and non-malignant conditions have reported an incidence of post-transplant IMC ranging between 1.5 and 22%. The median time to develop cytopenias was approximately 66 days to 40.2 months post-transplant (see Table 1) [1, 3•, 11–13, 14•, 15•, 16•, 17•, 18•]. Interestingly, very young infants (age < 3 months) who received human leukocyte antigen (HLA)-mismatched unrelated donor allogeneic HCT with umbilical cord blood cells were found to have a much higher incidence of IMC with rates of 44% at 1 year and 56% at 2 years post-HCT, and a median of 247 days [2].

There have been conflicting results regarding the significance of the conditioning regimen, patient age, transplant indication, or presence of GvHD in the development of IMC. However, most studies have consistently reported more frequent post-HCT IMC in younger patients, individuals transplanted for non-malignant conditions, and those receiving stem cells from an unrelated donor, particularly cord blood cells [1, 2, 3•, 11–13, 14•, 15•, 16•, 17•, 18•, 19]. Individuals with metabolic disorders, chronic granulomatous disease, and marrow failure conditions seem particularly at-risk [1, 2, 11, 12, 16•, 18•, 19]. Initial reports emphasized a high rate of mortality following the diagnosis of post-transplant hematologic autoimmunity, but with the development of new therapeutic interventions, mortality has greatly decreased with overall survival rates now exceeding 90% [1, 2, 3•, 4•, 14•, 12, 20]. Although IMC occur much more frequently following allogeneic HCT, they have additionally been reported following autologous HCT, particularly in patients with Hodgkin's disease [4•, 21–24].

Pathophysiology

The pathophysiologic mechanism behind the development of IMC in those who have received HCT remains unclear. However, it is likely due to multifactorial causes of immune dysregulation secondary to conditioning regimens, infectious insults, GvHD, immunosuppressive agents, and/or transfer of relatively naïve T lymphocytes and mismatched donor cells. The predominance in non-malignant disorders, often undergoing reduced intensity conditioned HCT, suggests a role for mixed donor chimerism and imperfect bidirectional tolerance. Functional regulatory T cells (Tregs) appear to play an important role in suppression of autoreactive T and B cells following the lymphopenic phase initially seen after allogeneic transplantation. This may be especially prominent in young infants with a paucity of Tregs within the developing thymus [2, 4•, 14•]. Interestingly, Kruizinga et al. identified a skewing towards a T helper cell type 2 (Th2) response in those with IMC post-HCT [16•]. Despite Tregs and Th2 cells arising from varying subsets of the CD4+ T cell lineage, they are intimately related and interact in the response to self-antigens with inadequate Tregs capabilities allowing for Th2-driven autoimmunity [25]. In those who undergo an autologous transplant, there also seems to be an imbalance of tolerance that allows autoreactive T cells to target self-antigens on hematopoietic cells [4•].

Immune dysfunction following transplantation is further exacerbated by immunosuppressive therapy for prevention of GvHD, particularly calcineurin inhibitors (CNIs), such as tacrolimus or cyclosporine. In studies evaluating immune-mediated disorders following solid-organ transplantation, CNIs are posited to contribute to IMC via decreased/dysfunctional Tregs and abnormal thymic T cell maturation [26, 27]. Post-HCT, these CNI immune alterations may be further exacerbated by radiation-induced thymic injury and/or impacts of conditioning serotherapy (anti-thymocyte globulin and alemtuzumab, for example) [2, 4•, 13, 16•, 19]. Interestingly, some cases of IMC arise or worsen after tapering of CNIs or other immune suppression for GvHD prevention or treatment, suggesting IMC as a form of GvHD. A role for mismatched antigens in the pathogenesis of alloimmunity following allogeneic transplant must be considered given the apparent increased risk for the finding in those receiving unrelated donor stem cells, particularly those receiving umbilical cords with greater mismatches than seen in marrow or peripheral blood stem cell products [2, 3•, 12, 13, 18•, 19, 28].

Viral infections, including cytomegalovirus (CMV), Epstein-Barr virus (EBV), and human herpesvirus-6 (HHV-6), have been associated with IMC post-HCT [16•, 29], similar to the association between preceding viral illness and hemolytic anemia or immune thrombocytopenia in healthy children [9]. The relatively naïve T cell content in umbilical cord blood (UCB) stem cell sources predispose recipients to a

Table 1 Significant studies analyzing immune-mediated cytopenias following hematopoietic cell transplantation

Reference	Cohort	Incidence of IMC	Risk factors	Survival	Treatment strategies
O'Brien et al. 2004 [1]	439 pediatric alloHCT patients in the United States	6% (1 year) cumulative incidence of AIHA	Age < 10 years and metabolic disorder	47%	Corticosteroids, IVIG, CSA discontinuation, Rituximab, Splenectomy, Mycophenolate, 2nd HCT, Plasmapheresis, EPO
Page et al. 2008 [2]	19 infant (≤ 3 months old) UCBT patients with metabolic or hemoglobinopathy disorders in the United States	44% (1 year) and 56% (3 years) cumulative incidence of IMC	Higher cumulative incidence of IMC with or without other manifestations of cGVHD	95%	Corticosteroids, IVIG, Rituximab, Azathioprine, CSA discontinuation, Splenectomy, EPO
Daikeler et al. 2013 [11]	726 UCBT patients in Europe	5% (1 year) and 6.6% (5 years) cumulative incidence of autoimmune diseases	Nonmalignant disease and interval from diagnosis to UCBT < 11.4 months	92.3% (for all post-UCBT autoimmune diseases)	Corticosteroids, Rituximab, CSA, Azathioprine, Mycophenolate, Tacrolimus, Plasma Exchange, Cyclophosphamide
Faraci et al. 2014 [12]	1574 pediatric alloHCT patients in Italy	2.1% incidence of IMC	Use of alternative donor and nonmalignant disease	85%	Corticosteroids, IVIG, Rituximab, Sirolimus, EPO, Plasma Exchange
Wang et al. 2015 [13]	533 adult alloHCT patients in the United Kingdom	3.6% incidence of AIHA	Unrelated donor and concordant gender between donor and recipient	64%	Prednisolone, IVIG, Rituximab, CSA, Mycophenolate, Sirolimus, Plasma Exchange, Azathioprine, Splenectomy
Chang et al. 2016 [14•]	265 pediatric alloHCT in Taiwan	6% incidence of AIHA	Thalassemia	80%	Methylprednisolone, IVIG, Rituximab, 6-Mercaptopurine
Bhatt et al. 2016 [3•]	152 HCT patients with hematologic malignancies the United States	7% at 3 years post day 100	No association between recipient age, diagnosis, conditioning intensity, ABO mismatch or recipient cytomegalovirus status	90%	Corticosteroids, Rituximab, IVIG, Splenectomy, CSA increase
Hwang-Bo et al. 2017 [15•]	292 pediatric alloHCT patients in South Korea	2.4% incidence of IMC	Unrelated donor and ATG conditioning exposure	100%	Corticosteroids, IVIG, Rituximab, Azathioprine, Mycophenolate, 6-Mercaptopurine, Splenectomy
Kruizinga et al. 2018 [16•]	531 pediatric alloHCT patients in the Netherlands	5.0% incidence of IMC	Cytomegalovirus reactivation, non-malignant disease, and pre-HCT alemtuzumab therapy	79%	Corticosteroids, IVIG, Rituximab, Bortezomib, Sirolimus, Splenectomy, Plasmapheresis, Stem cell boost
Gonzalez-Vicent et al. 2018 [17•]	4099 pediatric and adult alloHCT patients in Spain	1.5% incidence of AIHA	Age < 15 years, UCBT, and HLA mismatched donor	60%	Corticosteroids, IVIG, Rituximab, Mycophenolate, Cyclophosphamide, Alemtuzumab, Eculizumab, Bortezomib, Plasma Exchange, Splenectomy
Deambrosis et al. 2019 [18•]	36 UCBT patients with Hurler syndrome in the United Kingdom	22% incidence of IMC	Elevated ALC pre-transplant & fludarabine/ busulfan conditioning	87.5%	Prednisolone, Rituximab, Bortezomib, 2nd HCT, IVIG, Mycophenolate, Plasma Exchange, Vincristine, Cytoxan, Etanercept, Sirolimus

alloHCT allogeneic hematopoietic cell transplantation, *AIHA* autoimmune hemolytic anemia, *IVIG* intravenous immunoglobulin, *CSA* cyclosporine, *HCT* hematopoietic cell transplantation, *EPO* erythropoietin, *UCBT* umbilical cord blood transplantation, *IMC* immune-mediated cytopenias, *cGVHD* chronic graft-versus-host disease, *ATG* anti-thymocyte globulin, *ALC* absolute lymphocyte count

greater risk for viral reactivation, potentially contributing to increased IMC post-HCT [30]. Finally, it has been hypothesized that the increased risk of IMC post-HCT in non-malignant transplant recipients is secondary to the

relative competency and robustness of their immune systems, given a lack of previous exposure to cytotoxic or immunosuppressive agents [1, 18•].

Regardless of the initial triggering immunologic event, the cytopenias observed are due to auto- or allo-antibodies directly targeting hematopoietic cells. Antibodies directed against red blood cells may be categorized as "warm" or "cold" based on their reactivity at various temperatures. Those with warm antibodies present with immunoglobulin G (IgG) antibodies that optimally function at 37 °C while those with cold antibodies show immunoglobulin M (IgM) antibodies that react best at 4 °C, with additional functional abilities at 30–37 °C. Patients with warm antibodies may additionally show complement antibodies on serology (often C3) [9].

Warm autoantibodies can exhibit specificity for a particular blood group antigen, most frequently members of the Rhesus (Rh) blood group system, such as E or e [1, 2, 13, 16•]. There is sparse data regarding the identification of anti-platelet antibodies in IMC post-HCT, though the glycoprotein IIb/IIIa (GPIIb/IIIa) platelet-specific antigen is predominantly targeted in other settings of immune-mediated thrombocytopenia [9]. Various antibodies directed against the human neutrophil antigens (HNA) have been described in immune-mediated neutropenia post-HCT, but anti-HNA-2 antibodies are most prevalent [31, 32].

Once IgG or complement binds to the red blood cell surface, an interaction occurs with a Fc or complement receptor on a cytotoxic or phagocytic immune cell, resulting in extravascular hemolysis (typically splenic). A similar process results in the removal of targeted neutrophils and platelets. Direct destruction of the red blood cell by complement may also occur. IgM is especially efficient at initiating complement fixation and thus those with cold hemagglutinin hemolytic disease predominantly experience intravascular destruction of red blood cells. In addition to targeting circulating hematopoietic cells, autoantibodies also may directly target hematopoietic precursors within the bone marrow (see Fig. 1a) [9].

Clinical Manifestations and Diagnostic Testing

As previously mentioned, most large retrospective studies note the median time post-transplant to develop IMC to fall between 66 days and 40.2 months with patients capable of developing the condition despite receiving immunosuppressive therapy, including prophylaxis or treatment directed against GvHD [1, 3•, 11–13, 14•, 15•, 16•, 17•, 18•]. Hemolytic anemia is the most common cytopenia observed but immune-mediated thrombocytopenia and neutropenia can additionally occur, independently or in combination (see Table 2) [2, 3•, 4•, 11, 12, 15•, 16•, 18•, 29, 31].

In those presenting with hemolytic anemia, worsening pallor and fatigue can be observed with new onset jaundice, scleral icterus, dark urine, and splenomegaly. The degree of anemia is variable but hemoglobin drops can be significant (levels < 3 g/dL) and lead to fatal hemodynamic insufficiency if interventions are not initiated promptly [3]. Response to packed red blood cell transfusions is often poor or suboptimal, indicative of a peripheral destructive process. Laboratory evaluations may show elevated reticulocyte counts (though low if red blood cell progenitors are targeted), decreased haptoglobin levels, and increased indirect bilirubin/lactate dehydrogenase (LDH)/aspartate aminotransferase (AST) values. Care must be taken in the interpretation of haptoglobin values, as levels may be elevated during times of acute inflammation or low in young infants who often have poor synthesis capabilities for the protein. Urinalysis may show hemoglobinuria in cases of intravascular hemolysis. Examination of the peripheral blood smear reveals spherocytes, polychromasia due to reticulocytosis, and fragmented or nucleated red blood cells [9].

Direct Coombs testing often reveals complement (C3) and/or immunoglobulins (IgM or IgG) antibodies on the surface of red blood cells although a negative test does not rule out a diagnosis of immune-mediated hemolytic anemia. Indirect Coombs testing may also yield the presence of serum antibodies. It has also been observed that two distinct patient presentations may exist in those with hemolytic disease with patients having an early onset (2–8 months following transplant) often having a positive cold antibody and those with a positive warm antibody having a later onset of 6–18 months post-transplant [33].

Immune-mediated thrombocytopenia post-HCT may be isolated in nature or occur concurrently with hemolytic anemia (Evans syndrome) [3, 4•, 11, 12, 15•, 29]. Physical exam findings include increased/frequent bruising, petechiae, purpura (including oral "wet purpura"), menorrhagia, epistaxis, and gastrointestinal or urinary bleeding. Thrombocytopenia can be severe with platelet counts falling below 10×10^9/L though bleeding may be less severe overall compared to those with thrombocytopenia secondary to hypoproduction. A poor response to platelet transfusion can be observed and quantified by obtaining a platelet count 10–60 min post-transfusion (corrected count increment). Antiplatelet antibodies may be present on testing but their presence does not exclude other causes of thrombocytopenia and their absence does not rule out a diagnosis of an immune-mediated destructive process [9]. Immune-mediated neutropenia is the least common of the IMC post-HCT but has been reported [2, 11, 16•, 29, 31, 32]. Neutropenia can be severe (absolute neutrophil count < 500/μL), with variable response to granulocyte-colony stimulating factor [31, 32].

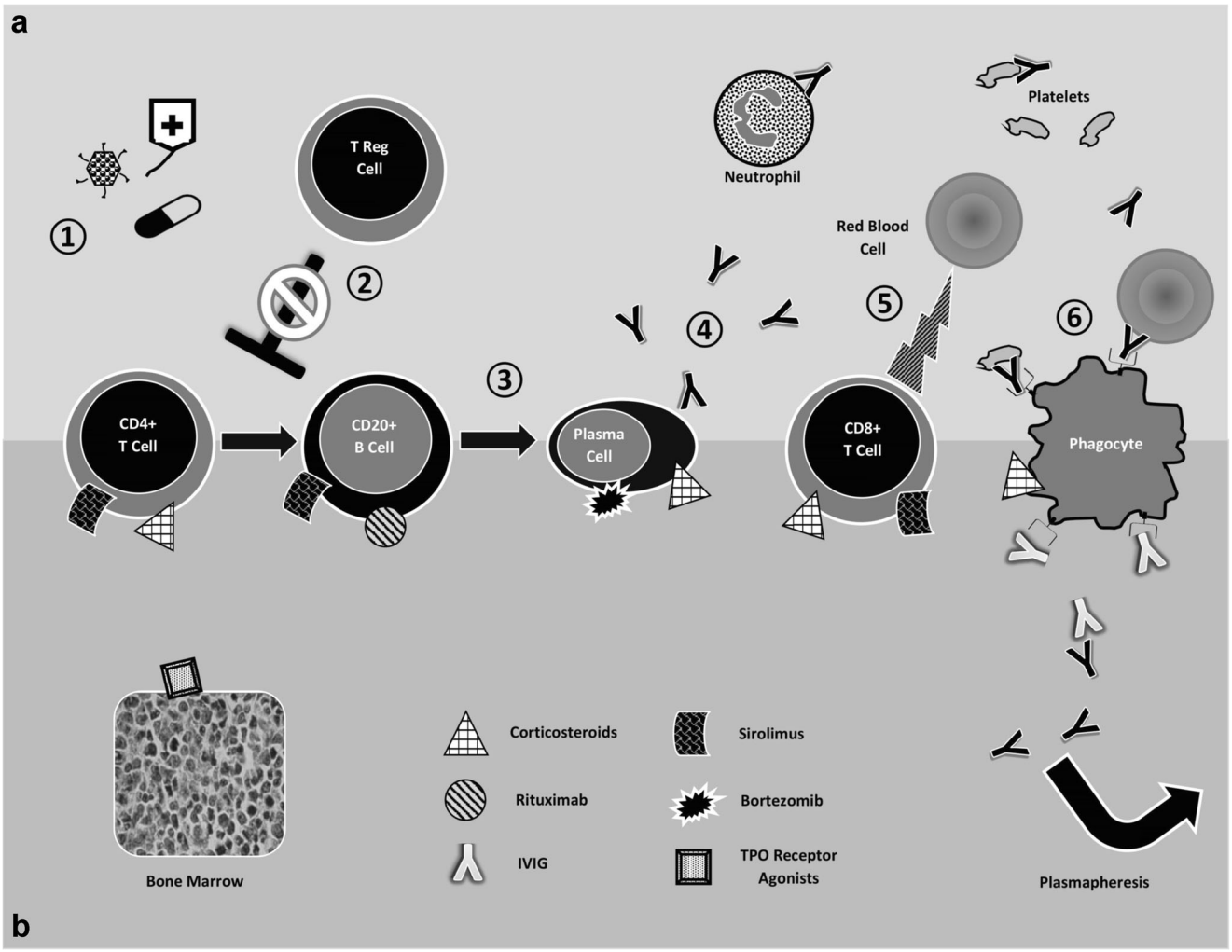

Fig. 1 Post-transplant immune-mediated cytopenias proposed pathophysiology and treatment strategies. **a** Potential pathophysiology of post-transplant IMC. (1) Immune dysregulation secondary to infectious insults, pre-transplant conditioning, and post-transplant immunosuppression. (2) Reduced and dysfunctional regulatory T cells (T regs) with an inability to suppress auto/alloreactive T and B cells. (3) B cells stimulated by auto/alloreactive CD4+ T helper cells transform into plasma cells. (4) Plasma cells produce antibodies targeting hematopoietic cells. (5) Direct damage to hematopoietic cells by CD8+ cytotoxic T cells may occur. (6) Hematopoietic cells coated with antibodies are cleared via phagocytes. **b** Treatment strategies for post-transplant IMC. High-dose corticosteroids decrease phagocytosis, reduce antibody production, and suppress T cell proliferation. Rituximab targets CD20+ B cells. Intravenous immunoglobulin (IVIG) blocks Fc receptors on phagocytes and directly inhibits antibodies preventing their binding to hematopoietic cells. Sirolimus hinders T and B cell activation and proliferation via mTOR inhibition. Bortezomib targets plasma cells through proteasome inhibition. Thrombopoiein (TPO) receptor agonists stimulate the production of hematopoietic cells via binding to hematopoietic stem cells and megakaryocytes within the bone marrow. Plasmapheresis directly removes circulating antibodies

Differential Diagnosis

Before confirming a diagnosis of an IMC, differential diagnoses for alternate post-HCT hematologic abnormalities should be considered and investigated (see Table 2). In-addition to primary or secondary graft failure, marrow suppression secondary to infection, and relapsed malignant disease, one should consider transplant-associated thrombotic microangiopathy (TA-TMA), drug-induced cytopenias, and ABO incompatibility leading to hemolytic anemia.

TA-TMA is a unique thrombotic microangiopathic hemolytic anemia caused by significant endothelial injury during HCT. Intensive conditioning, infection, immunosuppressive agents (e.g., calcineurin inhibitors), and GvHD all appear to play a role in vascular damage that leads to platelet activation, formation of microthrombi, and destruction of red blood cells, most prominently within the renal vasculature. Uncontrolled complement activation is also an important component of the disease pathogenesis [34]. Laboratory values are similar to those found in IMC with anemia, thrombocytopenia, elevated LDH, decreased haptoglobin, and schistocytes on peripheral blood smear, although an elevated creatinine, proteinuria, and increased terminal complement (soluble C5-b9) may aid in making the TA-TMA diagnosis. ADAMTS-13 levels are not

Table 2 Immune-mediated cytopenia evaluation

	Observation	Evaluation[a]	Treatment
Platelets	Unexplained decline in platelet count by ≥ 33% after platelet engraftment	Platelet Ab screen (for anti-HLA and anti-GP Abs)	Consider Ag neg, cross-matched or HLA-matched platelets
		Post-transfusion platelet count (10–60 min after transfusion) to calculate the corrected count increment	IMC treatment
	New transfusion requirement following platelet engraftment or platelet transfusion refractory	Consider non-immune causes of thrombocytopenia (DIC, sepsis, fever, bleeding, TA-TMA, sequestration) or drug induced immune thrombocytopenia	Discontinue/replace offending agent and monitor
	Failure to engraft platelets by day+ 60	Evaluate bone marrow for megakaryocytes and donor chimerism	Low megakaryocytes and expected donor chimerism
			➔ consider TPO agonist
			Low megakaryocytes and low donor chimerism
			➔ consider graft failure therapy
			Normal megakaryocytes ➔ IMC treatment
Neutrophils	Unexplained decline in absolute neutrophil count < 500	Neutrophil Ab screen	G-CSF 5 mcg/kg IV/SQ daily
			IMC treatment
		Consider non-immune causes of neutropenia (infection, medications)	Treat infection or discontinue/replace offending agent and monitor
RBCs	Unexplained decline in hgb ≥ 20%	Hemolysis screen: CBC, type & screen, DAT, reticulocyte count, peripheral blood smear, bilirubin, LDH, haptoglobin, UA	If Cold Hemagglutinin disease ➔ plasmapheresis and warming protocol for IVF/meds/ambient temperature
			If warm or mixed immune-mediated hemolytic anemia ➔ IMC treatment
	New pRBC transfusion requirement after achieving transfusion independence	Anti-ABO titer (if donor/recipient ABO incompatible), consider bone marrow evaluation for Pure Red Cell Aplasia	If Pure Red Cell Aplasia, consider weaning GvHD prophylaxis, steroids, IVIG, rituximab, bortezomib, erythropoietin, and/or plasmapheresis
		Consider non-immune causes of hemolysis: Hereditary RBC disorder, infection (Malaria, *C. perfringens*), DIC, mechanical (prosthetic heart valve, extracorporeal circuits), PNH, TA-TMA, hypersplenism, oxidant substances (dapsone), renal failure	Discontinue/replace offending agent and monitor, treatment specific to finding (ex. Eculizumab for TA-TMA)

[a] If IMC suspected in one cell line, recommend investigation of all three cell lines to guide IMC therapy if indicated

Ab antibody, *HLA* human-leukocyte antigen, *Ag* antigen, *IMC* immune-mediated cytopenias, *DIC* disseminated intravascular coagulation, *TA-TMA* transplant-associated thrombotic microangiopathy, *TPO* thrombopoietin, *G-CSF* granulocyte colony-stimulating factor, *RBCs* red blood cells, *hgb* hemoglobin, *CBC* complete blood count, *DAT* direct anti-globulin test, *LDH* lactate dehydrogenase, *UA* urinalysis, *pRBC* packed red blood cells, *PNH* paroxysmal nocturnal hemoglobinuria, *GvHD* graft-versus-host disease

normally decreased as in thrombotic thrombocytopenic purpura (TTP), and clinically, recalcitrant hypertension is also a prominent finding in TA-TMA. Most cases of TA-TMA occur within the first 100 days following transplant making its presentation typically earlier than most cases of IMC. Treatment involves withdrawal of calcineurin inhibitors and usage of the complement-blocking agent, eculizumab. Plasma exchange seems to be less effective than in TTP [34–36].

Donor-recipient red blood cell ABO incompatibility occurs frequently at the time of HCT, and thus, various hemolytic complications, including acute hemolysis, pure red cell aplasia, and delayed hemolysis secondary to passenger lymphocyte syndrome (PLS), may be observed by clinicians caring for transplant patients [36]. Major ABO incompatibility is seen most often in group O patients receiving stem cell products from group A, B, or AB donors although group AB donation to group A or B recipients can also lead to incompatibility reactions. Minor ABO incompatibility results from the transfer of donor plasma with high-titer isohemagglutinins to an incompatible transplant recipient and is more commonly seen in pediatric patients given their relative reduced blood volumes. Bidirectional incompatibility is defined as having both major and minor ABO mismatches between donor and recipient [36, 37]. Prevention can occur through depletion of donor red blood cells or plasma from the graft prior to infusion. Acute hemolysis with associated symptoms at the time of graft infusion is possible with major ABO incompatibility with pure red cell aplasia and delayed red blood cell engraftment becoming apparent later. Pure red cell aplasia will present with reticulocytopenia and the absence of erythroblasts on marrow evaluation [37, 38]. Minor ABO incompatibility can also manifest with sub-acute hemolysis of recipient red blood cells with additional delayed hemolysis occurring in the setting of PLS, through the process of transplanted donor B lymphocytes producing red blood cell-targeted antibodies. Both pathologies occur within the first 2 weeks post-transplantation and thus present much earlier than IMC post-HCT. Management involves blood product transfusion in severe cases with adequate fluid hydration during times of hemolysis. Pure red cell aplasia treatment is variable and frequently involves multiple treatment modalities, including corticosteroids, the anti-CD20 medication rituximab, and erythropoietin (EPO) [36–38].

Finally, pharmacologic agents leading to cytopenias through immune-mediated processes should be considered in the evaluation of a hematopoietic stem cell transplant patient presenting with worsening cytopenias. Anemia, thrombocytopenia, and neutropenia may all occur through drug-induced immunologic destruction. A thorough review of the patient's medications may reveal a likely drug culprit with special attention paid to antibiotics (e.g., cephalosporins, penicillins, vancomycin), rituximab, sulfa-containing medications, and CNIs. Cytopenias can be significant and treatment primarily involves withdrawal of the offending agent [36, 39, 40].

Treatment Strategies

Initial management in those with concern for an IMC post-HCT involves blood product support if hemodynamic instability is present and intravenous hydration during severe episodes of hemolysis. Therapies directed towards slowing the immunologic clearance of hematopoietic cells are similar to treatment interventions employed in previously healthy children presenting with immune-mediated cytopenias but post-HCT patients often require multiple therapies and have resistance to first-line management measures (see Table 3 and Fig. 1b) [4•, 12, 15•, 18•, 41].

First-line interventions include high-dose corticosteroids and intravenous immunoglobulins but given the fact that two-thirds of patients often fail to respond to these therapies, early initiation (within the first week of diagnosis) of rituximab should be strongly considered [2, 3•, 12, 13, 17•, 18•]. Various additional immunosuppressive therapies, including sirolimus, azathioprine, mycophenolate, and 6-mercaptopurine, have been used with varying success rates [1, 2, 16•, 14•, 42]. Bortezomib, a potent proteasome inhibitor, should be considered in those with particularly resistant disease, as the mechanism of action targeting plasma cell antibody production has been shown to be beneficial in those developing IMC post-HCT [19•, 18•, 43]. Use of the thrombopoietin agonists (e.g., romiplostim and eltrombopag) has also been reported to be successful in patients with persistent severe thrombocytopenia following transplant although response takes several weeks [29, 44, 45]. A trial of granulocyte-colony-stimulating factor (G-CSF) may be attempted in those with neutropenia but patients may not respond to this therapy [31, 32]. There are conflicting data on the benefits of escalation or reduction of CNIs in the setting of IMC post-HCT. When possible, CNIs should be rapidly weaned or replaced with alternative GvHD prophylaxis such as sirolimus. However, when IMC arises in the setting of a CNI wean, consideration for GvHD should be considered and the wean halted [1, 3•]. For patients with cold hemagglutinin disease, or IgM-mediated hemolytic anemia, plasmapheresis should be initiated immediately. Similarly, the highest risk population of patients, those < 3 years of age or post-HCT for inherited metabolic disorder, may benefit from up-front plasmapheresis as well, for direct removal of circulating antibody while additional medications address antibody production. Finally, there has not been a proven benefit to splenectomy, and given its associated risks, it is recommended to trial pharmacologic agents prior to spleen removal [1, 2, 3•, 17•, 16•].

Conclusions

Individuals undergoing allogeneic or autologous HCT are at risk for significant IMC. The pathogenesis of post-HCT IMC

Table 3 Immune-mediated cytopenia treatment algorithm by affected cell line

Isolated thrombocytopenia

1st line: IVIG 1 g/kg daily ×3, then consider weekly
2nd line: Add Methylprednisolone 2 mg/kg daily × 14, then taper over 8 weeks
3rd line: Add Rituximab 375 mg/m2 IV weekly, up to 4 doses (monitor IgG, may require IVIG replacement)
4th line: Alternative therapies

Isolated neutropenia

1st line: G-CSF 5 mcg/kg IV/SQ daily
2nd line: Add methylprednisolone 2 mg/kg daily × 14, then taper over 8 weeks, and IVIG 1 g/kg IV daily × 3, then consider weekly
3rd line: Add rituximab 375 mg/m^2 IV weekly, up to 4 doses (monitor IgG, may require IVIG replacement)
4th line: Alternative therapies

Hemolytic anemia in isolation or in combination with neutropenia or thrombocytopenia:

Standard risk (age > 3 years, no inherited metabolic disorder diagnosis):

1st line: ±G-CSF 5 mcg/kg IV/SQ daily prn neutropenia), and methylprednisolone 2 mg/kg daily × 14, then taper over 8 weeks, and rituximab 375 mg/m^2 IV weekly, up to 4 doses, and IVIG 1 g/kg IV daily × 3, then weekly
2nd line: plasmapheresis (single volume exchange every other day × 3–5 sessions), and bortezomib 1.3 mg/m^2 SQ twice weekly for total of 4 doses (on plasmapheresis days, give 2 h following plasmapheresis)
3rd line: Alternative therapies

High risk (age < 3 years and/or inherited metabolic disorder diagnosis):

1st line: ±G-CSF 5 mcg/kg IV/SQ daily prn neutropenia, and methylprednisolone 2 mg/kg daily × 14, then taper over 8 weeks, and plasmapheresis (single volume exchange every other day × 3–5 sessions), and bortezomib 1.3 mg/m^2 SQ twice weekly for total of 4 doses (on plasmapheresis days, give 2 h after completion of plasmapheresis), and rituximab 375 mg/m^2 IV weekly, up to 4 doses, and IVIG 1 g/kg IV daily × 3, then weekly
2nd line: Alternative therapies

Assessing response to therapy

Allow 2 weeks to determine response to a given lines of therapy, recognizing some interventions may take longer to have an impact (rituximab, bortezomib for example) whereas others are much faster-acting (methylprednisolone, IVIG, plasmapheresis).
In general, treatment is considered successful and safe to taper when ANC exceeds 500, pRBC transfusions < every 7 days and platelets transfusions < every 3 days.

Alternative therapies

Mycophenolate mofetil, azathioprine, vincristine, cyclophosphamide, daratumumab, splenectomy

Guidance of GvHD prophylaxis

Controversial given timing of IMC and individual patient clinical picture. Calcineurin inhibitors have been associated with IMC. Consider weaning off and/or replacing with sirolimus. However, if IMC occurred in midst of immunosuppression taper, consider possible overlap with GvHD, and holding taper or returning to previous dose.

Supportive care interventions

Treatment	Risks	Consideration
Corticosteroid	Opportunistic infections	Anti-fungal / anti-bacterial / anti-viral prophylaxis and surveillance
	Adrenal insufficiency	Stress dose steroids with procedures, illness; ACTH stimulation test
	Hypertension	Monitor and treat
	Gastritis	Protein-pump inhibitor or H2 blocker
	Bone demineralization	Vitamin D and calcium monitoring and supplementation
	Insomnia/agitation	Dose corticosteroids QAM, sleep hygiene, olanzapine prn
	Infection with immunizations	Avoid live vaccines
Rituximab	Risk of hepatitis B activation	Assess patient serologies
	B cell aplasia / hypogammaglobulinemia	IVIG supplementation for IgG <400
	Inadequate immunization response	Delay immunizations until 6 months after rituximab
Splenectomy	Infection with encapsulated organisms	Penicillin prophylaxis if <5 years of age, immunize against *H. influenzae, N. meningitidis, S. pneumoniae* (ideally 4 weeks prior to splenectomy)

is multifactorial (i.e., immune dysregulation with autoantibody production following the conditioning regimen, infections, GvHD, immunosuppression agents, and/or decreased/dysfunctional Tregs and mismatched naïve T cell containing stem cell infusions). Patients at greatest risk for developing post-transplant IMC include very young infants (age < 3 months) and individuals receiving transplant for non-malignant conditions, particularly inherited metabolic disorders, and/or from unrelated umbilical cord blood cells. Various conditions can lead to post-transplant hematologic dysfunction and should be considered prior to making a diagnosis of IMC. Management typically involves multiple agents targeting antibody production. Studies have shown that early induction of rituximab is associated with sustained remissions and encouraging survival outcomes.

Compliance with Ethical Standards

Conflict of Interest Thomas F. Michniacki, Christen L. Ebens, and Sung Won Choi declare they have no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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