Biol Blood Marrow Transplant 22 (2016) 1552–1564

Biology of Blood and Marrow Transplantation

journal homepage: www.bbmt.org

ASBMT American Society for Blood and Marrow Transplantation

The Role of Biomarkers in the Diagnosis and Risk Stratification of Acute Graft-versus-Host Disease: A Systematic Review

Alaa M. Ali, John F. DiPersio, Mark A. Schroeder*

Washington University School of Medicine, Department of Medicine, St. Louis, Missouri

Article history:
Received 21 December 2015
Accepted 26 April 2016

Key Words:
Graft-versus-host disease
Hematopoietic stem cell transplantation
Biomarker
Proteomics

A B S T R A C T

Allogeneic hematopoietic cell transplantation (HCT) is an increasingly used curative modality for hematologic malignancies and other benign conditions. Attempts to reduce morbidity and mortality and improve survival in patients undergoing HCT are crucial. The ability to diagnose acute graft-versus-host disease (aGVHD) in a timely manner, or to even predict aGVHD before clinical manifestations, along with the accurate stratification of these patients, are critical steps to improve the treatment and outcomes of these patients. Many novel biomarkers that may help achieve these goals have been studied recently. This overview is intended to assist clinicians and investigators by providing a comprehensive review and analytical interpretation of the current knowledge concerning aGVHD and biomarkers likely to prove useful in diagnosis and risk stratification of this condition, along with the difficulties that hamper this approach.

INTRODUCTION

Acute graft-versus-host disease (aGVHD) is a major complication of allogeneic hematopoietic cell transplantation (allo-HCT), occurring in up to 60% of transplant recipients, with well-described risk factors, including HLA mismatch, age, stem cell source, donor–recipient sex disparity, and conditioning regimen [1]. aGVHD is a systemic disorder driven by donor T cells with a pleomorphic clinical presentation involving multiple target organs, including the skin, liver, and gastrointestinal (GI) tract [2]. The diagnosis of aGVHD with clinical and pathological confirmation is helpful but lacks positive predictive value (PPV) and negative predictive value (NPV) [3-5].

Immunosuppressive therapy with steroids is the first-line therapy for clinically significant graft-versus-host disease (GVHD). Mortality and morbidity from high-dose systemic immunosuppression is significant, and no other treatment added to upfront steroids has proven beneficial to date [6].

Although the clinical diagnosis can be readily made in patients with a classical presentation, some cases prove challenging [7]. Currently, the diagnosis of aGVHD can be made by combining the clinical impression, high pretest likelihood of aGVHD, exclusion of other competing disorders, along with histological examination of the target tissue. Biopsy alone is not the gold standard for diagnosis, owing to the false-negative results (patchiness of the disease, absence of the typical changes at early stages) and false-positive results (residual conditioning regimen toxicity, infections) [3-5]. Furthermore, in many patients other causes of symptoms may be found, confounding the diagnosis of aGVHD [8-12].

Once diagnosed, the severity of aGvHD has historically been graded using the Keystone Consensus criteria [13] or CIBMTR criteria [14]. Grading was primarily developed as an important tool to determine the appropriate management of aGvHD and to assess the response to therapy. Grading is also important due to its impact upon survival and association with graft-versus-leukemia effect. It has been well recognized that current grading systems overgrade some patients with a high likelihood of responding to immunosuppressive therapy and can not predict who will respond to steroids with certainty [15]. There are several limitations to current prognostic grading systems: despite the general relationship to outcomes, inter-observer errors can occur due to subjective biases, initial grade may not reflect peak grade and the time to response after therapy initiation is not accounted for. Therefore, many patients who are classified as standard-risk have their treatment fail while others classified as high- risk are over-treated. Recently, a refined clinical grading system was introduced as a better tool not only to stratify patient's

Financial disclosure: See Acknowledgments on page 1562.

* Correspondence and reprint requests: Mark A. Schroeder, MD, Division of Oncology, Washington University School of Medicine, 660 S Euclid Ave, Campus Box 8007, St. Louis, MO 63110.
E-mail address: mschroed@dom.wustl.edu (M.A. Schroeder).

http://dx.doi.org/10.1016/j.bbmt.2016.04.022
1083-8791/

non-relapse mortality risk but also predict response to therapy with high risk patients less likely to respond to steroids compared to standard risk patients [15]. However, this refined scoring system could still be plagued by inter-observer biases with significant variability across BMT centers [16]. It is hoped that the use of biomarkers can add to prediction accuracy and eliminate some of these problems.

High-dose systemic steroids are the standard first-line therapy for patients with grade II or higher aGVHD [6,17]; however, practices differ among institutions with respect to initial steroid dose, the use of additional immunosuppressive agents, and the approach to steroid tapering after initial response. In general, therapy of clinically suspected aGVHD is started before diagnosis is confirmed and potentially before peak grade is achieved. Such an approach may expose the already immunosuppressed patients to unnecessary systemic steroids, with significant infectious and noninfectious complications. In fact, a study conducted to reduce the dose of initial steroid therapy for grade I-II acute GVHD found no difference in outcomes between standard dose (2 mg/kg) and reduced dose (1 mg/kg) arms [18]. Approximately one-half of patients have a complete response to steroids by day 28 after therapy initiation [19].

Steroid-refractory aGVHD is associated with high transplantation-related mortality (TRM) and low overall survival, even without relapse of the underlying disease that necessitated HCT [20]. There are no standard second-line therapies for steroid-refractory aGVHD, and responses vary, but based on small retrospective and phase I/II studies, responses are around 30% to 50% with a 6-month NRM of approximately 50% [6]. A biomarker-based grading system may have the potential to more accurately stratify patients based on risk of failure to respond to steroids or alternative treatment approaches, and may be used to help identify additional lines of therapy in those that fail upfront therapy.

Although mortality related to GVHD has been reduced in recent years [21,22], aGVHD remains a major cause of TRM. aGVHD represents the primary limitation to more widespread use of allogeneic HCT as a potentially curative modality for patients with malignant and nonmalignant diseases. The field of biomarker research may provide more accurate grading/risk stratification and identification of patients at greater risk for refractoriness to therapy or GVHD progression. Furthermore, the treatment of aGVHD has recently evolved from a one-size-fits-all approach to a more refined strategy based on predicted outcomes. Patients who are predicted to have low-risk aGVHD may benefit from lower doses and shorter courses of immune suppression. In addition, because not all cases of aGVHD progress in the same way or have the same outcome, the therapy should be tailored not only to the severity of the disease, but also to the predicted rate of progression. As a result, numerous researchers have examined whether adding novel plasma biomarkers at different time points before and after transplantation can add to the accuracy of prediction compared with other prognostic tools. Timely recognition of patients who are at high risk for aGVHD or who would likely demonstrate resistance to steroids early in the course of transplantation may lead to more stringent monitoring, better preventive care, and introduction of alternative and more effective immunosuppressive treatments earlier in the course of treatment.

It is reasonable to assume that plasma proteins involved in the complex pathophysiology of aGVHD might be altered in these patients. For the past 20 years, various groups have been investigating potential biomarkers to enhance the early and more accurate diagnosis and risk stratification of patients with aGVHD. Recent research has applied proteomics technologies to identify aGVHD biomarkers. This has led, in a short period, to the identification of novel biological pathways and biomarkers predictive of and associated with aGVHD [23]. Nevertheless, no single biomarker or panel of biomarkers has been validated for clinical use via large multicenter trials. In this article, we summarize the current knowledge of promising diagnostic and prognostic aGVHD biomarkers and analyze the supporting data available in the literature.

REVIEW DESIGN

We searched PubMed and MEDLINE up to December 31, 2015, to identify studies evaluating biomarkers in the setting of aGVHD. Each biomarker (ie, micro RNA [miRNA], ST2, TNF receptor 1 [TNFR1], IL-7, sBAFF, REG3α, S100, TIM-3, CK-18, hepatocyte growth factor [HGF], and elafin) was searched separately as well. Only full-text articles published in English were considered. The primary search was conducted using the terms “graft-versus-host disease” and “biomarker,” excluding reviews. Relevant references in the publications identified were reviewed as well. Eligible studies included clinical studies with more than 5 patients. Studies investigating the diagnostic and prognostic value of transcriptomic and proteomic biomarkers were reviewed. Here we discuss biomarkers that were evaluated in at least 2 independent studies. The primary statistical outcomes were sensitivity, specificity, PPV, NPV, area under the curve (AUC), and hazard ratio (HR). The main outcomes of the remaining preclinical and clinical studies were reported in a table but not discussed in the text.

DIAGNOSTIC AND PROGNOSTIC BIOMARKERS

The biomarkers of aGVHD that have been discovered so far can be classified in various ways, including target (systemic or particular organ/tissue) and biophysical properties. Systemic biomarkers lack organ specificity for skin or GI aGVHD. These biomarkers rise in response to systemic injury rather than to specific tissue damage. On the other hand, organ-specific biomarkers are expressed by target organs rather than the effector cells that are damaging all tissues. Identifying markers that are target tissue specific has been technically challenging, owing to the cellular heterogeneity of tissues and the difficulty of amplifying the amount of protein required.

Another way of classifying these novel biomarkers is based on their biophysical properties. Transcriptomic biomarkers are discovered by RNA expression profiling (mRNA, rRNA, tRNA, and other noncoding RNAs), whereas proteomic biomarkers are discovered by the methodical study of the protein profile of a biologic specimen. Finally, cellular biomarkers are discovered by the studying of the altered numbers and functions of several different immune cell subsets [24-26].

Below we review the systemic biomarkers (miRNA, ST2, and markers of immune activation) and organ-specific biomarkers (Reg3α, S100, TIM-3, CK-18, HGF, and elafin). Our findings are summarized in Table 1.

SYSTEMIC BIOMARKERS

miRNAs

miRNAs are a class of small noncoding RNAs (21 to 25 nucleotides) that negatively and positively regulate gene expression by translational repression or induction of alterations in messenger RNA stability. miRNAs regulate gene function in various ways and at multiple levels, particularly