

Meeting Report

The Role of Donor-Specific HLA Alloantibodies in Liver Transplantation

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The impact of donor-specific HLA alloantibodies (DSA) on short- and long-term liver transplant outcome is not clearly defined. While it is clear that not all levels of allosensitization produce overt clinical injury, and that liver allografts possess some degree of alloantibody resistance, alloantibody-mediated adverse consequences are increasingly being recognized. To better define the current state of this topic, we assembled experts to provide insights, explore controversies and develop recommendations for future research on the consequences of DSA in liver transplantation. This article summarizes the proceedings of this inaugural meeting.

Several insights emerged. Acute antibody-mediated rejection (AMR), although rarely diagnosed, is increasingly understood to overlap with T cell-mediated rejection. Isolated liver allograft recipients are at increased risk of early allograft immunologic injury when preformed DSA are high titer and persist posttransplantation. Persons who undergo simultaneous liver–kidney transplantation are at risk of renal AMR when Class II DSA persist posttransplantation. Other under-appreciated DSA associations include ductopenia and fibrosis, plasma cell hepatitis, biliary strictures and accelerated fibrosis associated with recurrent liver disease. Standardized DSA testing and diagnostic criteria for both acute and chronic AMR are needed to distil existing associations into etiological processes in order to develop responsive therapeutic strategies.

Keywords: Antibody-mediated rejection, donor-specific HLA antibodies, graft outcomes, liver transplant, renal transplant, simultaneous liver–kidney transplant

Abbreviations: ADCC, antibody-dependent cellular cytotoxicity; AMR, antibody-mediated rejection; DSA, donor-specific HLA alloantibodies; HCV, hepatitis C virus; MFI, mean fluorescence intensity; NK, natural killer; SLKT, simultaneous liver–kidney transplantation; SPI, solid-phase immunoassay

Received 09 September 2013, revised 23 December 2013 and accepted for publication 13 January 2014

Introduction

Advances in intensive care, immunosuppressive medications and chronic disease management have dramatically improved short- and intermediate-term liver allograft survival. Nonetheless, the 10-year survival rate, based on the Scientific Registry of Transplant Recipients, is only 54% and roughly mirrors that for kidney transplants. Late-onset morbidity and mortality are often attributed to cardiovascular complications, recurrent liver disease and malignancy; however, with better recognition and control of recurrent disease, an accurate assessment is needed to quantify the potential adverse impact of donor-specific HLA alloantibodies (DSA) on liver transplant outcomes. Even though the detrimental effects of DSA on outcomes following renal

transplantation have been recognized since 1969, our understanding of their pathologic impact continues to evolve (1). It is clearly demonstrated that other solid organ allografts are adversely impacted by DSA, and that kidney allograft survival is longest among patients without DSA. Thus, as a general matter of alloimmune biology, there is legitimate precedent for concern in the liver transplant realm.

Until recently, preformed DSA were generally considered to be clinically irrelevant to liver allograft outcomes (2–5) based on the perceived absence of hyperacute rejection and the rarity of early allograft loss from rejection (6). However, adequately powered studies since the early 1990s have shown that liver transplant recipients with a positive crossmatch have an increased risk of early allograft damage and failure (7). Recent studies have confirmed inferior clinical outcomes in some but not all DSA-positive patients (2–4,8), and it seems prudent to reexamine the impact of DSA on liver allograft structure and function, including long-term survival. Toward this goal, highly regarded experts in histocompatibility, nephrology, hepatology, immunology, pathology and transplant surgery met to review and discuss the current literature and paradigms regarding the role of DSA in liver transplantation (Appendix). The attendees of this meeting were tasked to translate what has been learned about diagnosis, treatment and outcomes in renal and liver transplantation into a consensus view of current practice, and identify goals for research on DSA in liver transplantation that will facilitate advancement of our understanding of the field.

Current State-of-the-Art Testing Techniques

Since the landmark study of Patel and Terasaki in renal transplants in 1969 (1), DSA testing has been mandated prior to renal allograft transplantation, and the technology to detect DSA has advanced dramatically. First employed was the cytotoxic crossmatch, which has limited sensitivity and specificity, could not differentiate IgG from IgM antibodies, did not distinguish HLA from non-HLA antibodies, required an adequate supply of viable lymphocytes and resulted in suboptimal virtual crossmatching. Despite its limitations, cytotoxic crossmatching performed pretransplantation essentially eliminated hyperacute renal allograft rejection. This technology has since been supplemented by flow cytometry and solid-phase immunoassays (SPIs).

Although flow cytometric crossmatching facilitates identification of HLA and some non-HLA DSA without dependence on complement, it lacks sufficient sensitivity and specificity to characterize all HLA alloantibodies present. By contrast, multi-analyte bead assays performed on the Luminex[®] platform allow for highly accurate characterization of HLA alloantibodies present at very low levels, thereby allowing assessment of both the level and temporal endurance of

DSA. However, the high sensitivity of SPIs increases their susceptibility to environmental variables and interference by substances in serum (9). For these reasons, a cell-based assay is frequently paired with an SPI that utilizes purified Class I and/or II HLA antigens as targets, collectively enabling detection of HLA alloantibodies with high specificity and sensitivity. The concurrent development of polymerase chain reaction–based molecular methodologies has optimized HLA typing and identification of HLA antigens/alleles. A multiplexing fluorescence-based assay allows the simultaneous detection of up to 100 uniquely coated microparticles (10).

Nonetheless, the role of SPIs in kidney transplantation remains controversial: while some investigators have reported that antibodies encountered exclusively by SPI are associated with a significant risk of early antibody-mediated rejection (AMR) and graft loss (11), other investigators conclude that such antibodies are clinically irrelevant (12). The different conclusions are likely explained by differing patient factors and/or methodological or technological aspects of SPI testing. Clinical factors accounting for the different conclusions include differences in patient populations (e.g. primary vs. regrant, male:female ratio, presence or absence of prior sensitization), the immunosuppression regimen, the performance or lack of performance of protocol biopsies and assessment of different end points. Technical factors include: (1) the mean fluorescence intensity (MFI) cutoffs that each laboratory establishes to assign antibodies (13); (2) the antigen source (native or recombinant); (3) the conformation of the antigen (intact or denatured); (4) antigen density; (5) the presence or absence of interfering factors; and (6) various biologic aspects of HLA alloantibodies (e.g. complement activation, immunoglobulin subclass, common epitopes shared between unique antigens). Resolving variability among centers requires recognition that SPI and MFI metrics are not quantifiable measures of antibody (i.e. MFI is not a titer). It is critical that manufacturers minimize lot-to-lot variability and improve the quality of target molecules conjugated to microparticles. This will allow for inter-center optimization and standardization of protocols, which is both feasible and imperative for the future (13).

There is great interest in developing molecular platforms to diagnose AMR in serum, urine or allograft tissue. These efforts are progressing rapidly (14) and are currently available for use in clinical trials. Early efforts in renal allografts have focused on endothelial activation (14), which will have to be validated at multiple centers and then tested separately in liver allografts. Since numerous studies have underscored the major role of nonadherence to immunosuppression, efforts to prevent and detect such nonadherence should be incorporated into the design of future studies (8,15). In addition, molecular phenotyping may help to discern critical differences between patients with DSA in serum who are: (1) not experiencing pathologic injury; (2)

experiencing covert pathologic injury that only later becomes clinically apparent; and (3) experiencing overt pathologic injury (16).

Acute Antibody-Mediated Liver Allograft Rejection

Many patients with low-to-moderate MFI Class I DSA have no apparent quantifiable consequences of DSA in the absence of recurrent disease, but the full spectrum of potential insults associated with some DSA has yet to be identified (Table 1). The concepts of acute AMR (3) occurring early after transplantation and chronic (or indolent) AMR occurring late after transplant as broad categorizations of antibody-mediated injury are gaining traction in the literature. Although hyperacute rejection has been reported (6), the more common and most widely recognized presentation attributed to acute AMR is otherwise unexplained liver allograft dysfunction, falling platelet and complement levels and increased levels of circulating immune complexes in patients with preformed persistent DSA in whom liver biopsy shows microvascular injury in addition to other characteristics commonly associated with allograft rejection (17). This occurs most commonly during the first several weeks after transplantation and is seen in approximately 1% of all early (<90 days) liver allograft failures, but the rate increases to 10% of idiopathic early liver allograft failures in DSA-positive patients (18).

Animal models have provided insights into the relative resistance of the liver to acute antibody-mediated injury, as well as identification of factors that override these natural defense mechanisms. Mechanisms of resistance to acute antibody-mediated injury include: the large size and unconventional sinusoidal microvascular bed of the liver; secretion of high levels of soluble HLA antigens; Kupffer cell phagocytosis of immune complexes, platelet aggre-

gates and activated complement components; the dual afferent circulation of the liver; a homologous complement source; and the ability of the liver to repair and regenerate after injury. Conversely, high-level sensitization; IgG antibodies, especially those subclasses that efficiently fix complement and mediate antibody-dependent cellular cytotoxicity (ADCC); reduced-size grafts; Kupffer cell inactivation or depletion; and antibody targeting of endothelial cell antigens all increase susceptibility to AMR-related injury. Acute AMR has been produced reliably in highly sensitized rats and confirmed in rhesus macaques. In the original rat model, antibody-mediated injury was characterized by congestion, edema and hemorrhage into the liver parenchyma (19) and passive administration of HLA Class I antibodies, alone, in high titers was sufficient to cause injury. Revival of interest in animal models of liver allograft AMR will be essential to a better understanding of: (1) all the possible acute and chronic consequences of DSA and association of damage or accommodation with antibody class, subclass, MFI and specificity; (2) characteristics and timing of ADCC pathways; (3) endothelial physiologic consequences of antibody binding; and (4) potential novel therapeutic strategies.

Other Manifestations of DSA in Liver Transplant Recipients

Focus on acute AMR is attributable to its known association with alloantibodies and poor outcome in highly sensitized and suboptimally treated recipients. Emerging evidence, however, points toward the possibility of a broad range of potential DSA-associated pathologies that all require further study, preferably as part of prospective clinical trials (Table 1). The relative contribution of antibodies to graft injury has been difficult to tease out because: (1) rejection, currently categorized as “cellular,” may represent a mixture of T cell and antibody-mediated changes; (2) the hepatic sinusoidal microvasculature is normally lined by natural killer (NK) cells, NK T cells and macrophages (Kupffer cells), whereas in other organs the presence of these cells is associated with ADCC mechanisms; and (3) some changes likely evolve over years to decades.

Regardless, indication liver biopsy specimens from DSA-positive recipients are more likely to demonstrate rejection and steroid-resistant rejection when C4d tissue staining is found (20), patients with preformed Class II DSA are at higher risk of early liver allograft rejection (21) and patients with DSA posttransplantation have an increased risk of chronic rejection (22,23), especially when treated with less aggressive immunosuppressive regimens.

Little data exist on the effect of chronic DSA and potential long-term consequences of immunosuppression minimization in their presence. Several, but not all (16), studies in pediatric liver transplant patients show an association between DSA and progressive fibrosis (24), while other

Table 1: Potential associations of donor-specific HLA alloantibodies with outcomes in liver transplant or simultaneous liver–kidney transplant recipients

(1) Hyperacute rejection
(2) Acute antibody-mediated rejection ¹
(3) Early acute “cellular” rejection
(4) Steroid-resistant rejection
(5) Chronic rejection
(6) Plasma cell (<i>de novo</i> autoimmune) hepatitis
(7) Idiopathic fibrosis progression
(8) Accelerated fibrosis in hepatitis C viremic patients
(9) Anastomotic biliary strictures
(10) Portal venopathy and nodular regenerative hyperplasia
(11) Antibody-mediated renal allograft rejection in simultaneous liver–kidney transplant recipients ¹

No associations have been confirmed in large randomized controlled trials.

¹Consensus opinion of experts agrees on these associations based on multiple case–control studies.

studies suggest that DSA IgG subclass might be important in distinguishing pathologic versus nonpathologic DSA (Kaneku H, et al, manuscript in preparation). Further investigation is needed to determine the relative contributions of HLA and non-HLA DSA, technical complications and abnormal graft physiology to determine the underlying cause(s) of injury. However, ultimately we may need to redefine outcomes of interest; although death and advanced fibrosis are clearly the most important outcomes, more subtle possible effects of DSA on hepatic stellate cells and liver sinusoidal endothelial cells need exploration. In fact, one focus of future research should be to understand how the effects of DSA vary depending on cofactors, some of which may promote immunostimulatory/profibrogenic effects while others could promote toleragenic effects. The contribution of cofactors, such as up-regulation of intra-graft HLA antigens by recurrent disease, must also receive attention. For example, inflammatory-mediated tissue damage up-regulates targets of DSA in allografts and is associated with fibrosis progression in hepatitis C virus (HCV) viremic patients. Although causation has not been established, multivariable analysis in a single-center retrospective study found preformed DSA to be an independent predictor of fibrosis progression after controlling for potential confounders (25). In contrast, Tregitopes may be formed as a result of DSA, thereby promoting tolerance (26). Once phenotypes are better characterized, there will be opportunity to uncover key factors involved in the evolution of these disparate pathways.

Biliary strictures have also been seen more commonly in patients with DSA than in those without DSA. In fact, the two factors independently associated with biliary anastomotic strictures are ABO-incompatibility and the presence of Class II DSA (27).

Plasma cell (*de novo* autoimmune) hepatitis is a rare but well-described cause of liver allograft injury and dysfunction associated with both DSA and autoantibodies. In pediatric liver allograft recipients, DSA were significantly more common in patients with plasma cell hepatitis than in either tolerant or nontolerant patients without plasma cell hepatitis (28). Moreover, the pattern of histopathologic injury is similar to autoimmune hepatitis and suggests an overlap between allo- and auto-immunity, perhaps related to epitope spreading and development of DSA.

In essence, better characterization of DSA and allograft phenotypes is needed. Further advances in our understanding of the full spectrum of AMR will depend on more precise characterization of DSA and cross-platform analyses that include routine and multiplex protein immunohistochemistry, messenger RNA and microRNA expression arrays, proteomics and metabolomics with attention to changes in endothelial cell physiology and microvascular architecture.

DSA in Simultaneous Liver–Kidney Transplant Recipients

Recognition of the inability of the liver to afford renal allografts complete protection from preformed DSA has been described previously, but despite a 17% 6-month renal allograft survival in crossmatch-positive patients in 1994 (29), allocation policies have not been revised to account for this risk. More specific DSA determinations with SPI led to the discovery that renal allograft protection by the liver allograft occurs when the recipient harbors isolated preformed Class I DSA in low-to-moderate amounts (Table 2). Protection is incomplete, however, in recipients with preformed Class II DSA, in which case both the kidney and liver allografts are at risk for rejection (Figure 1) (21), especially when Class II DSA persist posttransplantation (30). Analyses that lack differentiation between Class I and II DSA or those without a focus on posttransplant persistence of DSA may be underpowered to find these associations (31,32). Fortunately, the risk of persistence is low and at least partly dependent on class and MFI: Class I with MFI $\geq 5000 = 5\%$, Class II with MFI 5000–9999 = 23% and Class II with MFI $\geq 10\,000 = 33\%$ (4).

Importantly, antibody binding is largely stochastic and influenced by antigen and antibody density. Class I HLA is constitutively expressed on all cells within the liver, whereas Class II HLA is limited largely to subsets of resident hematolymphoid cells in normal livers. Inflammatory, infectious or ischemic injury can up-regulate Class II HLA within the liver, including the portal capillary microvascular endothelia, thereby resulting in variable antigen density (33). As with other organs (but more pronounced because of the liver's size), secretion of soluble HLA Class I and II antigens can neutralize DSA by forming immune complexes that are cleared by Kupffer cells, thereby augmenting antibody-mediated injury. The renal peritubular capillary bed is one-hundredth (0.21 m^2) that of the liver (21 m^2) in area, and the concentration of bound antibody within the liver allograft is therefore diluted. Although the size of the liver may mitigate the effect of preformed DSA, the impact is likely spectral.

Despite a lack of randomized controlled trials, the available literature consistently documents inferior outcomes in patients who undergo simultaneous liver–kidney transplantation (SLKT) when high MFI Class II DSA is present (21,29). Because the most effective treatment of AMR is avoidance, patients who undergo SLKT should ideally receive organs without Class II antigens against which the recipient has DSA with an MFI $>10\,000$. However, the risks of waiting versus the risks of proceeding must always be critically evaluated in an era when the average Model for End-Stage Liver Disease score at transplantation is continually increasing. When patients must receive crossmatch-positive organs, postoperative testing to determine persistence of antibody and close follow-up with a low

Table 2: Possible reasons why the liver can withstand and, in some cases, protect other organs from donor-specific HLA alloantibodies (DSA)

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- (1) Soluble Classes I and II antigen secretion is increased during injury
 - (2) Increased area of distribution
 - a. Liver capillary surface area (21 m²) is 100 times that of the kidney (0.21 m²)
 - b. HLA Class I antigens are present on all liver cells
 - (3) Dual blood supply of the liver: reciprocal blood flow regulation facilitates improved blood flow during injury. This may decrease hepatic necrosis from arterial vasospasm and local intrahepatic coagulation that can occur as a result of DSA
 - (4) Hepatocyte regenerative capacity is tremendous
 - (5) Impaired coagulation may facilitate continued blood flow during antibody-mediated rejection (AMR)
 - a. Thrombocytopenia: most liver transplant candidates have portal hypertension-induced thrombocytopenia. Because platelet aggregates are an integral part of DSA-induced vascular thrombosis, thrombocytopenia may decrease injury
 - b. Coagulopathy: patients with either liver dysfunction (decreased production) or cholestasis (decreased vitamin K absorption) frequently have coagulopathy
 - (6) Hypocomplementemia
 - a. Cirrhotic patients have lower C3, C4 and CH50 levels
 - b. Hepatitis C viral proteins may inhibit C3 production
 - (7) Kupffer cells
 - a. Facilitate removal of platelet aggregates, immune complexes and activated complement that are produced during AMR
 - b. Activated Kupffer cells can promote immune reactivity of lymphocytes and coagulation
 - (8) Antigen location and density and DSA concentration and characteristics
 - a. HLA Class I is expressed on all cells in the liver and at high density on all endothelial cells
 - b. HLA Class II is constitutively expressed only on dendritic cells and some macrophages and in a minority of individuals on the portal microvasculature
 - c. HLA Class II antigen expression is up-regulated in the setting of inflammation, infectious and ischemic injuries on the portal microvascular, sinusoidal and central vein endothelium, bile ducts and hepatocytes
 - d. Time since sensitization impacts antibody concentration
 - e. Antibody characteristics, including class and subclass, Fc binding, complement fixation and antigen affinity, likely affect their injurious potential
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threshold for allograft biopsy seems prudent, although the precise testing interval and threshold for biopsy are not yet determined.

Pathology Resulting From DSA in Liver Transplant Recipients

The hepatic microvasculature of the liver differs substantially from that of other solid organs; the one conventional capillary bed in the liver is the peribiliary capillary plexus. The larger hepatic microvasculature, including the sinusoids, is normally lined by macrophages (Kupffer cells), NK cells and NK T cells, whereas in kidney allografts, monocytes/macrophages and NK cells facilitate Fc-receptor-mediated recruitment and endothelial cell injury. During the first month after transplantation, AMR induces microvascular injury and a spectrum of findings that closely mimic preservation/reperfusion injury. More severe manifestations of early acute injury include platelet margination, arterial vasospasm, cellular recruitment and duct injury (17,34), with frequent evolution to histopathologic features traditionally attributed to cellular rejection; isolated AMR, in the absence of features consistent with acute cellular rejection, is rare.

The combination of AMR and T cell-mediated rejection includes microvascular endothelial cell hypertrophy and microvasculitis, resulting in eventual destruction, which, in

turn, leads to ductopenia related to destruction of the peribiliary plexus and direct biliary damage (35). Current stringent criteria for the diagnosis of acute AMR in liver allograft recipients include: (1) DSA in serum; (2) histopathologic evidence of diffuse microvascular endothelial cell injury and microvasculitis; (3) strong and diffuse C4d positivity in tissue; and (4) reasonable exclusion of other causes of injury that might result in similar findings (18). The granularity of these criteria will increase over time. For example, the presence of C4d staining is most convincing of antibody involvement when the staining is diffuse, and detection of C4d is more sensitive in fresh tissue, although formalin-fixed tissue can be used (3,36). Like renal allografts, the pathology associated with C4d-negative AMR following liver transplantation will need to be elucidated, and other, more robust, markers of antibody-mediated endothelial cell injury need to be developed.

Potential features of chronic AMR are beginning to appear in the literature but need to be further substantiated. Included are subsinusoidal and perivenular fibrosis, which has been associated with DSA and C4d staining. Endothelial cell reactivity and microvasculitis have not been described. Experience with this form of injury, however, is limited to a few studies, established diagnostic criteria do not yet exist and the pattern(s) of injury will have to be distinguished from technical complications and other causes of a similar pattern of fibrosis, such as altered allograft physiology.

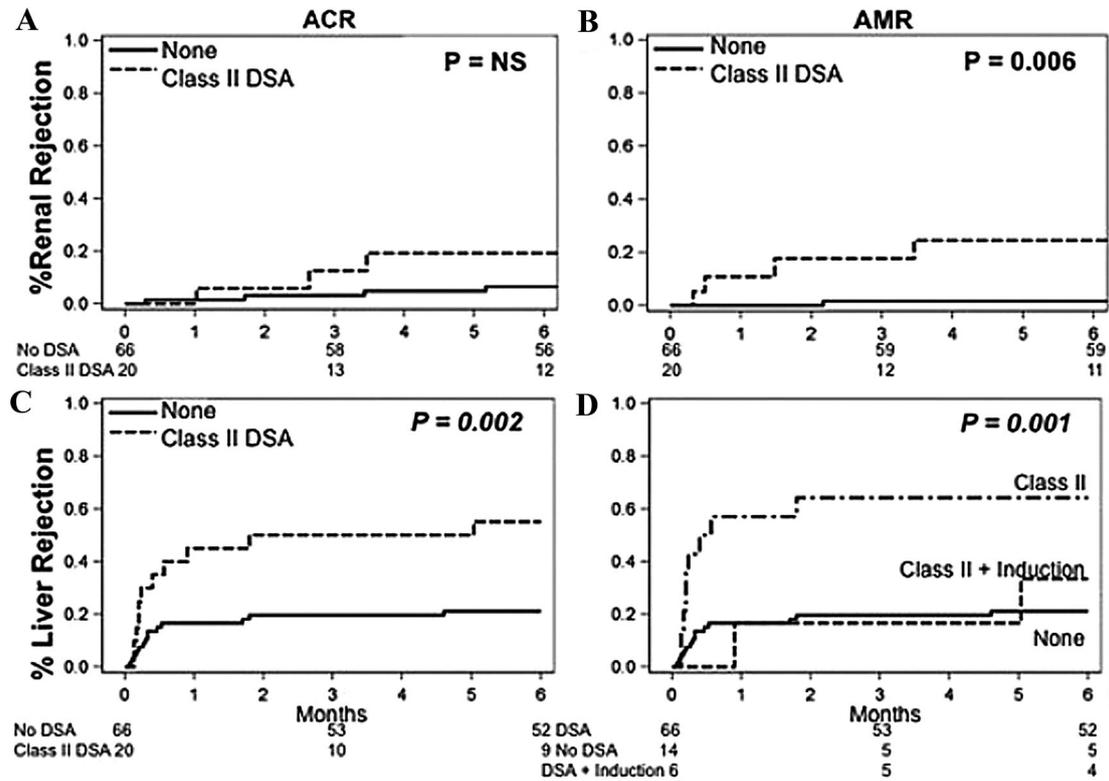


Figure 1: The presence of Class II donor-specific HLA alloantibodies (DSA) in serum (A) did not change the risk of renal acute cellular rejection (ACR) and (B) increased the risk of renal acute antibody-mediated rejection (AMR). The increased risk (C) of liver allograft rejection was abated with (D) antibody induction, but this did not improve the overall impaired survival. (Reproduced with permission: O'Leary JG, et al *American Journal of Transplantation* 2013 (21).)

Insights Into Monitoring

Ideally all liver allograft recipients would be tested for DSA pretransplantation, and positive patients retested 1–2 weeks posttransplantation to determine persistence. However, most patients with preformed low-to-moderate levels of isolated Class I DSA in the absence of recurrent liver disease appear to have few, if any, short- or long-term consequences. Nevertheless, sufficient evidence has accrued to show that acute antibody-mediated injury in liver transplant and SLKT recipients is a potential cause of occasionally serious allograft injury. The challenge, therefore, is to develop a cost-effective DSA monitoring algorithm that: (1) reliably identifies the small percentage of sensitized patients before transplantation who are likely to experience severe adverse consequences of DSA early after transplantation; and (2) reliably identifies DSA characteristics late after transplantation that signal inadequate immunosuppression or an unacceptable risk of chronic allograft injury (5,8).

Monitoring for anti-donor HLA Class II antibodies might be particularly relevant since their low expression in the graft does not result in their effective removal from the circulation, probably for the reasons cited above (37). These

antibodies can increase the risk of early renal and liver allograft rejection and death (4,21,29). Patients with preformed DSA may benefit from closer monitoring and a lower threshold for allograft biopsy in the presence of dysfunction. The significance of DSA late after transplantation without allograft dysfunction is uncertain but in isolation is not an indication for intervention. The long-term outcomes of such patients are as yet unknown and need further evaluation. Future research, therefore, is warranted to determine the most cost-effective approach to pretransplantation and follow-up testing. In addition, thresholds for concern, allograft biopsy and therapy all need to be identified in prospective clinical trials.

Insights into treatment

Individual anti-humoral agents and techniques used in renal transplant recipients (plasmapheresis, intravenous immune globulin, rituximab, bortezomib and eculizumab) are most commonly employed as multimodality regimens, making the relative contribution of the component therapies difficult to ascertain. The published literature on utilization of anti-humoral therapies in liver transplantation is quite limited. Bortezomib is a proteasome inhibitor with potent immunomodulatory properties that has been used to treat

acute AMR in liver allograft recipients (38). Its immunomodulatory properties include: (1) induction of endoplasmic reticulum stress and apoptosis in plasma cells; (2) induction of cell cycle arrest in T and B lymphocytes and (3) inhibition of nuclear factor kappa-B-mediated B cell and plasma cell survival. Important considerations exist regarding anti-humoral therapies in liver transplant recipients because of their potent immunosuppressive effects that may exacerbate chronic viral hepatitis or increase infectious risks. Therefore, the combination of avoidance/prevention when possible may be the best strategy. Toward this goal, *de novo* DSA prevention strategies include strict adherence to immunosuppression and use of tacrolimus (rather than cyclosporine) (8). However, when prevention fails, prospective treatment trials are needed to determine the safest and most effective treatment for acute AMR in liver allograft recipients. Prospective clinical trial design will require the careful selection of end points.

these associations require confirmation in prospective clinical trials. Effective therapeutic strategies cannot be developed until granular clinicopathologic criteria for diagnosis and end points for study are established. Criteria for acute AMR are nearing consensus agreement, but detection of chronic AMR will likely require additional studies. Even then, caution will be needed in patients with viral infections. Fortunately, acute AMR is uncommon, but other less profound pathologic consequences of DSA likely occur. Evidence of an association between DSA and these other potential pathologic manifestations is mounting, but causation has not been established. Likely, not all DSA are pathogenic, and antibody class, amount, specificity, functional properties, epitope binding, and antigen density and location determine the ultimate outcome; however, the competing theory that all DSA are pathogenic, although some consequences may be subclinical or may develop over long periods of time, will need to be definitely disproven in large, long-term prospective trials. Ultimately, the sensitivity and specificity of testing must improve the correlation with clinical outcomes, perhaps as a result of analysis of antibody characteristics, greater understanding of the pathophysiology of injury, isolation of donor and/or recipient phenotype or genotype characteristics, improved molecular diagnostics or simply the bundling of several

Conclusions

Antibodies have been associated with a spectrum of injury in liver allografts that is probably more common than previously appreciated (Table 1; Figure 2), although many of

<p>Facts:</p> <ul style="list-style-type: none"> ▪ Antibody-mediated rejection (AMR) occurs in liver allografts. ▪ The liver does not completely protect the kidney from donor-specific HLA alloantibodies (DSA) in recipients of a simultaneous liver–kidney transplant. ▪ Currently available testing has dramatically improved over the past few decades, but sensitivity and specificity still vary depending on the outcome considered.
<p>Possibilities:</p> <ul style="list-style-type: none"> ▪ Do only some or do all DSA eventually lead to pathologic outcomes? ▪ Does DSA cause or accelerate fibrosis progression in liver allografts with or without recurrent disease? ▪ Are their cofactors (or synergizers) that enhance the injurious potential of DSA? ▪ In patients with preformed DSA what role does cellular memory play in outcome? ▪ Is DSA associated with or causative of plasma cell (<i>de novo</i> autoimmune) hepatitis? ▪ What precise role does DSA play in rejection, steroid resistant rejection and chronic rejection?
<p>Opportunities:</p> <ul style="list-style-type: none"> ▪ Define the pathologic criteria to diagnose AMR in liver allografts. ▪ Improve or replace C4d staining in formalin-fixed liver tissue. ▪ Characterize the molecular signature of AMR in liver transplantation. ▪ Exploit animal models of liver AMR to define resistance mechanisms and characterize acute and chronic consequences of DSA. ▪ Characterize the following cohorts of patients with preformed DSA: <ul style="list-style-type: none"> • Those who will benefit from altered immunosuppression. • Those who can be successfully treated. • Those who must be listed with for liver transplantation with unacceptable antigens. ▪ Develop a monitoring schedule for DSA post-liver transplantation. ▪ Determine if all DSA are eventually pathologic over decades of exposure. ▪ Design therapeutic AMR treatment trials in liver allograft recipients.

Figure 2: Conclusions from the meeting.

tests. In the absence of the consummate test, we must strive to mitigate the effects of DSA on liver allografts by determining which patients: (1) will not develop clinically significant long-term consequences; (2) will benefit from altered immunosuppression regimens; (3) can be treated successfully; and (4) should be listed for transplantation with unacceptable antigens. Given the potential acute and/or chronic consequences of DSA, once the highest risk groups can be reliably identified, the decision to deal with the subsequent consequences will need to be addressed on a per patient basis; the days of complete disregard for DSA in liver transplantation are drawing to a close.

Acknowledgments

We gratefully acknowledge the unrestricted gifts/grants from the Baylor Health Care System Foundation, Novartis, Thermo Fisher Scientific, Astellas, Genentech and Texas Transplantation Society that made this conference possible.

Disclosure

The authors of this manuscript have conflicts of interest to disclose as described by the *American Journal of Transplantation*. P.I. Terasaki was the Chairman and a major investor in One Lambda, and P.F. Halloran owns shares in Transcriptome Sciences, Inc. All other authors have no conflicts of interest to disclose.

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Appendix

Faculty at the 2013 Antibody-Mediated Rejection in Liver Transplantation meeting.

R.Y. Calne, R. Colvin, A.J. Demetris, S. Feng, L.S. Friedman, H.M. Gebel, P.F. Halloran, A.D. Kirk, G.B. Klintmalm, S.J. Knechtle, S.V. McDiarmid, L.B. Melton, J.G. O’Leary, A. Shaked, P. Stastny, P.I. Terasaki, A. Thompson, K.J. Tinckam, S.J. Tomlanovich, J.F. Trotter, F. Vincenti, K.J. Wood, E.S. Woodle, A.A. Zachary.