



Review Article

Copyright © All rights are reserved by Jonathan RT Lakey

Alloimmune Recognition and Repeat Dosing of Allogeneic Mesenchymal Stromal Cells

Jonathan RT Lakey^{1,2*}, Carolina Blüguermann^{2,3}, Krista Casazza^{2,3}, Michael Alexander¹, Waldemar Learnhardt², Brad Robinson², and Adrian A Mutto^{2,3}

¹University of California, Irvine, Irvine, CA, USA

²Cellarion Lab, Irvine, CA, USA

³Laboratorio de Biotecnologías Aplicadas a la Reproducción y Mejoramiento Genético Animal, Instituto de Investigaciones Biotecnológicas IIBIO Dr. Rodolfo Ugalde, Universidad Nacional de San Martín, Argentina

***Corresponding author:** Jonathan RT Lakey, PhD, MSM, Professor Emeritus, Departments of Surgery and Biomedical Engineering, University of California, Irvine, CA, USA.

To Cite This article: Jonathan RT Lakey*, Carolina Blüguermann, Krista Casazza, Michael Alexander¹ Waldemar Learnhardt, Brad Robinson, and Adrian A Mutto, Alloimmune Recognition and Repeat Dosing of Allogeneic Mesenchymal Stromal Cells. *Am J Biomed Sci & Res.* 2026 32(4) AJBSR.MS.ID.004179, DOI: [10.34297/AJBSR.2026.32.004179](https://doi.org/10.34297/AJBSR.2026.32.004179)

Received: 📅 October 10, 2026 **Published:** 📅 October 16, 2026

Abstract

Allogeneic Mesenchymal Stromal Cells (MSCs) are often selected for their manufacturing scalability and immunomodulatory properties, but their description as absolutely immune privileged is unsupported. Animal studies demonstrate cellular and humoral responses to mismatched MSCs, and human studies document treatment-emergent anti-human leukocyte antigen antibodies, including donor-specific antibodies. These findings establish immunogenic potential but do not demonstrate universal rejection or a progressive decline in efficacy with repeated administration. This narrative review distinguishes immune recognition, sensitization, cell clearance, and clinical consequences; evaluates mechanistic, preclinical, and human evidence; and proposes a framework for repeat-dose development. Interpretation requires attention to donor identity, recipient immune status, administration route, manufacturing attributes, assay methods, and follow-up. Cord-tissue products require the same product-specific assessment as adult-tissue products. Importantly, clearance may contribute to immunomodulation through apoptosis and efferocytosis, so persistence alone is an insufficient surrogate for therapeutic activity. Repeat-dose studies should link serial immune measurements to subsequent-dose pharmacodynamic and clinical outcomes. The priorities are reliable attribution of sensitization, determination of its functional consequences, and identification of circumstances in which donor selection or regimen modification improves benefit and reduces risk.

Keywords: Mesenchymal stromal cells, Allogeneic cell therapy, Immunogenicity, Donor-specific antibodies, HLA, Immune privilege, Repeat dosing, Umbilical cord, Wharton's jelly

Introduction

Mesenchymal Stromal Cells (MSCs) have been investigated across inflammatory, immune-mediated, degenerative, and regenerative indications because of their immunomodulatory, trophic, angiogenic, and tissue-reparative properties [1]. Clinical develop

ment of the therapeutic potential of MSCs has been accelerated on part, by the feasibility of manufacturing capabilities. A key advancement in MSC therapeutics has been the development of allogeneic products from expanded donor-derived cell banks, creating the

prospect of standardized, inventory-based therapy rather than patient-specific manufacturing [2]. A central biological premise supporting this model was the characterization of MSCs as “immune privileged.” Early in vitro experimentation showed relatively low expression of Major Histocompatibility Complex (MHC)/Human Leukocyte Antigen (HLA) class I, limited constitutive HLA class II and costimulatory-molecule expression, and weak stimulation in mixed lymphocyte assays [3]. These observations were interpreted to mean that MSCs could cross histocompatibility barriers with little meaningful immune recognition [4,5]. However, animal studies subsequently demonstrated cellular and humoral responses to MHC-mismatched MSCs, including memory responses and rejection of allogeneic cells, shifting the field toward the more qualified concepts of “immune evasive,” “hypoimmunogenic,” or “relatively low immunogenicity” [6,7]. Human studies have reinforced the need for this distinction by documenting anti-HLA and Donor-Specific Antibody (DSA) formation after allogeneic MSC administration [8,9]. An equally important correction is required in the opposite direction, i.e., evidence of immune recognition is not synonymous with clinically important graft rejection, treatment failure, or cumulative toxicity [10,11]. In a systematic assessment of clinical alloimmune responses available to date, DSA formation was observed in a minority of treated patients and was not clearly associated with reduced safety or tolerability [12]. This distinction matters most when allogeneic MSCs are administered repeatedly. Re-exposure to mismatched donor antigens provides a biologically plausible setting for immune priming, memory, accelerated clearance, or altered therapeutic activity [13,14]. Preclinical models support this concern, but human evidence remains heterogeneous and insufficient to conclude that repeated dosing universally causes progressive loss of efficacy [15,16]. The question is particularly relevant to umbilical cord- and Wharton’s jelly-derived products, which are often positioned as low-immunogenicity sources and may be used in repeated-dose protocols. The appropriate translational framework must therefore separate four related but non-equivalent phenomena: immune recognition, sensitization, cell clearance, and clinically consequential loss of safety or efficacy. This narrative review examines the evidence that challenged the immune-privilege paradigm, evaluates what human data do and do not establish about alloimmunization and repeat dosing, and proposes practical immune-monitoring and trial-design considerations for contemporary allogeneic MSC development.

Review Approach

This narrative review draws on the supplied evidence summary and a targeted literature update and reference verification completed on October 3, 2026. Searches of PubMed-indexed records and publisher pages combined terms for mesenchymal stromal or stem cells with allogeneic, immunogenicity, alloimmunization, donor-specific antibodies, repeat dosing, umbilical cord, apoptosis, and efferocytosis. Primary human studies reporting immune mea-

surements were prioritized; foundational animal experiments were retained for mechanistic interpretation, and recent perspectives were used to frame research questions. Findings were compared by tissue source, donor relationship, recipient immune status, route, dosing schedule, assay, and follow-up. No quantitative pooling, formal risk-of-bias scoring, or exhaustive systematic search was performed.

From Immune Privilege to Conditional Immune Evasion

MSCs also exert broad immunomodulatory effects through soluble mediators and cell-cell interactions [17]. Although the inherent properties of MSCs in theory reduce the intensity of allorecognition relative to more immunogenic cell types, this state of reduced immunogenicity exists under conditions rather than an intrinsic absence of antigenicity [18]. Inflammatory licensing, differentiation, culture conditions, passage history, cryopreservation, tissue source, and local microenvironment can alter the immunologic phenotype of MSCs [19]. HLA expressions can increase under inflammatory conditions, and differentiated progeny may not retain the same immunologic profile as the administered stromal-cell product. [7,13] Accordingly, the immune phenotype relevant to clinical use is dynamic and product specific. The phrase “immune privileged” is therefore problematic because it collapses lower immunogenicity into immune invisibility.

Preclinical Evidence Establishes Immunogenic Potential

Several foundational in vivo studies directly challenged the assumption that allogeneic MSCs are intrinsically tolerated. Eliopoulos and colleagues demonstrated rejection of allogeneic marrow stromal cells in MHC-mismatched mice [20]. Nauta and colleagues showed that donor-derived MSCs could induce a memory T-cell response and promote donor graft rejection in an allogeneic host [21]. Badillo and colleagues subsequently demonstrated both cellular and humoral alloimmune responses to murine bone-marrow stromal progenitor cells. [6] Together, these studies established a principle that remains important for clinical translation: low stimulatory activity in vitro does not reliably predict absence of immune recognition in vivo. Preclinical repeat-dose studies further suggest that a first exposure can alter the response to subsequent allogeneic MSC administration. In an equine model, a second intra-articular injection of allogeneic MSCs produced a greater inflammatory response than a second autologous injection, consistent with acquired immune recognition [22]. Differentiation can also increase immunogenicity and constrain durability; in cardiac repair models, differentiation-associated increases in MHC expression were associated with immune rejection and reduced long-term benefit of allogeneic MSCs [23]. These studies establish biological plausibility for sensitization-dependent loss of persistence or activity. They should not, however, be treated as direct evidence that the same magnitude or clinical consequence occurs in humans.

Human Alloimmune Evidence

In a systematic review including 555 participants, of whom 356 received allogeneic MSCs treatment-emergent DSA was identified in a subset of recipients without a clear association with reported safety or tolerability [12]. This was not an adequately powered pooled test of efficacy loss after successive doses. Variation in immune testing and clinical context limits interpretation of negative associations. In a Crohn disease sub study, local administration of allogeneic adipose-derived MSCs generated DSA in some recipients, with pre-existing immunity influencing subsequent sensitization. Antibody levels decreased during follow-up in some groups, and investigators did not detect an association between DSA generation and the measured therapeutic response [16]. These findings concern a specific product and local administration regimen; they cannot establish the effect of repeated intravenous dosing. Sun and colleagues treated 12 children with autism spectrum disorder using one, two, or three intravenous cord-tissue MSC doses at two-month intervals. Five of nine initially anti-HLA-negative participants developed class I antibodies; no antibody-associated clinical manifestations were reported [24]. The mixed dosing cohorts, small sample and uncontrolled design do not establish a dose-dependent sensitization rate or preserved efficacy after sensitization. A 2024 reanalysis of two kidney-transplant MSC cohorts illustrates the difficulty of attributing antibody specificity. High-resolution HLA information changed interpretation of some previously assigned DSA, and shared donor mismatches complicated separation of responses to the kidney graft and MSCs [25]. Therefore, an antibody detected after MSC administration is not automatically caused by MSC exposure, particularly when recipients have additional allogeneic exposures. In a 2025 kidney-transplant study, six human participants were enrolled and four received 12 monthly kidney-donor-derived MSC infusions with concurrent immunosuppression. Infusions were acutely tolerated, but immune tolerance was not achieved, and DSA or rejection affected eligibility for or success of immunosuppression withdrawal [26]. The shared organ/MSC donor and changing immunosuppression prevent attribution of these outcomes to MSC sensitization alone. A 2025 single-dose lumbar facet study enrolled ten patients and reported no clinical signs of an immune reaction [27]. Its report did not include serial anti-HLA or DSA outcomes. Clinical tolerability without immune testing therefore cannot establish absence of sensitization.

Cord Tissue and Wharton Jelly Products

Umbilical cord tissue and Wharton's jelly are source descriptors, not interchangeable standardized products. The cord-tissue findings above should not be generalized to every isolation method, tissue compartment, donor bank, or manufacturing process. Claims of lower immunogenicity require comparisons of final products under comparable conditions. Tissue origin alone is insufficient to justify omission of immunogenicity assessment.

Clearance and Therapeutic Activity

Cell loss is not necessarily therapeutic failure. Pang and colleagues showed that blocking MSC apoptosis attenuated immunosuppression in experimental disease models and implicated efferocytosis, the phagocytic removal of apoptotic cells, in the host response [28]. These findings establish a mechanism in the tested models, not a universal requirement for every MSC product. Alloantibody-mediated clearance could reduce viable-cell exposure, alter host-mediated activity, or have little effect; the direction must be measured. A 2026 expert perspective emphasizes functional persistence and delivery context rather than cell tracking alone, [29] also highlighted in a recent review [30], supporting pairing viable-cell measurements with host-response biomarkers.

Repeat Dose Development

A test of immune-mediated loss of response requires temporally ordered measurements: immune status before each administration, product exposure and early pharmacodynamic activity after dosing, and clinical outcomes at prespecified intervals. Analyses should relate acquired immunity to the response to a subsequent dose, account for donor and lot changes, and distinguish loss of initial benefit from disease progression or variation in manufacturing potency. Missing immune samples and withdrawals after reactions should be reported because they can bias later-dose comparisons. Table 1 presents a proposed framework, not a universal regulatory requirement. Donor typing and serial serology are especially informative in repeat-dose programs where sensitization may affect retreatment or future transplantation. Cellular assays, functional antibody testing, and cell tracking should be selected according to mechanism, analytical feasibility, and the clinical question (Table 1).

Table 1: Proposed assessments for repeat-dose allogeneic MSC studies.

Domain	Assessment And Timing	Interpretation
Donor and recipient	Type donor HLA sufficiently to assign specificity; retain donor and lot identities for every dose.	Record relevant recipient typing and pregnancy, transfusion or transplant exposures.
Baseline and retreatment	Collect serum before treatment and, when feasible, before subsequent administrations.	Separate pre-existing sensitization from newly detected or amplified responses.
Postdose serology	Prespecify post dose and end-of-course sampling, with later follow-up for persistence; identify missing samples.	Report class I/II specificity, DSA denominator, assay platform, threshold and signal; do not equate mean fluorescence intensity with antibody titer.

Functional immunity	Use selected donor-reactive cellular or antibody-functional assays in mechanistic sub studies.	Serology alone cannot measure all cellular responses or prove cytotoxicity.
Final product	Document source/compartiment, passage, post-thaw viability, potency, HLA phenotype and process materials.	Assess batch effects separately from recipient sensitization.
Regimen and outcomes	Record dose, route, interval and concomitant therapy; link immunity to the next dose pharmacodynamic and clinical response.	Use prespecified temporal comparisons and report uncertainty rather than inferring causality from seroconversion.
Future transplantation	For relevant populations, characterize persistent antibody specificity with transplant specialists.	Potential future donor incompatibility is distinct from demonstrated MSC-related clinical harm.

Clinical and Regulatory Context

Approved repeat-dose therapy demonstrates that allogeneic MSC development need not depend on an absolute immune-privilege claim. The US prescribing information for remestemcel-L-rknd recommends eight initial intravenous infusions for pediatric steroid-refractory acute graft-versus-host disease, with additional treatment determined by response [31]. This establishes an indication-specific clinical use, not freedom from alloimmunization or applicability to immunocompetent recipients and other products. Persistent antibodies could affect future donor compatibility if their specificities overlap those of a later organ or hematopoietic graft. The kidney-transplant reanalysis shows why specificity and competing exposures require careful attribution [25]. This is a rationale for follow-up in transplant-relevant populations, not evidence that MSC administration has reduced subsequent transplant access.

Evidence Gaps and Limitations

The principal evidence gap is prospective linkage of acquired donor-reactive immunity to the response to a later MSC dose. Priority studies should incorporate adequate donor typing, distinguish same donor redosing from donor switching, and report product attributes and concomitant therapies. Source comparisons require comparable manufacturing and delivery conditions; otherwise, apparent source effects may reflect differences in the administered products. This synthesis is limited by targeted rather than exhaustive retrieval and the absence of formal risk-of-bias assessment. Small clinical cohorts, inconsistent immune ascertainment, missing samples, limited follow-up and recipient immunosuppression constrain estimates of incidence and outcome associations. Transplant recipients may have multiple sources of alloantigen exposure, and local administration cannot be assumed to predict intravenous responses. No universal sensitization rate, clinically validated antibody threshold, or causal estimate of repeat-dose efficacy loss can be derived from this evidence.

Conclusions

Allogeneic MSCs have context-dependent immunogenicity. Clinical development should determine whether donor-reactive immunity changes subsequent-dose activity or safety, using assays capa-

ble of assigning specificity and outcomes aligned with the intended mechanism. Manufacturing accessibility and immune compatibility are complementary development considerations. Product-specific evidence, rather than a categorical immune-privilege label, should guide donor selection, monitoring, and retreatment.

Declarations

Funding: No specific funding was received for this work.

Author contributions: Jonathan R T Lakey: Conceptualization, supervision, writing; review and editing. Carolina Blüguermann: Methodology, investigation, writing; review and editing. Krista Casazza: Conceptualization, methodology, investigation, writing; original draft, writing; review and editing. Michael Alexander: Investigation, writing; review and editing. Waldemar Learnhardt: Writing; review and editing. Brad Robinson: Project administration, writing; review and editing. Adrian A. Mutto: Conceptualization, supervision, writing; review and editing. All authors meet the journal's authorship criteria, have reviewed and approved the final manuscript, agree to its submission, and accept accountability for the work Ethics approval: Not applicable. This review reports no new human or animal research.

Data availability: No new participant-level datasets were generated. Evidence is available in the cited publications.

Acknowledgements

None.

Conflicts of Interest

All authors are employed by or serve as consultants to Cellarion. These relationships represent potential conflicts of interest. The authors are responsible for the interpretation of the evidence and the conclusions presented in this review.

References

1. Nurlybek T, Altaeva N, Kazhiyakhmetova B, Zhansaya Seitkumarova, Yerkezhan Baidildina, et al. (2026) Immunomodulatory Mechanisms of Mesenchymal Stromal Cells: Cytokine Networks and Therapeutic Potential Across Immune-Mediated, Inflammatory, and Regenerative Disorders. *Biology* 15(10): 794.

2. Karantalis V, Schulman IH, Balkan W, Hare JM (2015) Allogeneic cell therapy: a new paradigm in therapeutics. *Circ Res* 116(1): 12-15.
3. De Miguel MP, Fuentes Julián S, Blázquez Martínez A, CY Pascual, MA Aller, et al. (2012) Immunosuppressive properties of mesenchymal stem cells: advances and applications. *Curr Mol Med* 12(5): 574-591.
4. Dominici M, Le Blanc K, Mueller I, Fc Marini, Ds Krause, et al. (2006) Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy* 8(4): 315-317.
5. Ankrum JA, Ong JF, Karp JM (2014) Mesenchymal stem cells: immune evasive, not immune privileged. *Nat Biotechnol* 32(3): 252-260.
6. Cequier A, Vázquez FJ, Vitoria A (2024) The systemic cellular immune response against allogeneic mesenchymal stem cells is influenced by inflammation, differentiation and MHC compatibility: in vivo study in the horse. *Front Vet Sci* (11): 1391872.
7. Berglund AK, Fortier LA, Antczak DF, Schnabel LV (2017) Immunoprivileged no more: measuring the immunogenicity of allogeneic adult mesenchymal stem cells. *Stem Cell Res Ther* 8(1): 288.
8. Morin Zorman S, Loiseau P, Taupin JL, Caillat Zucman S (2016) Donor-Specific Anti-HLA Antibodies in Allogeneic Hematopoietic Stem Cell Transplantation. *Front Immunol* (7): 307.
9. Gladstone DE, Bettinotti MP (2017) HLA donor-specific antibodies in allogeneic hematopoietic stem cell transplantation: challenges and opportunities. *Hematol Am Soc Hematol Educ Program* 2017(1): 645-650.
10. Arabi TZ, Almasry Y, Xue A (2025) Immune rejection of human mesenchymal stem cells compared to extracellular vesicles in mice with renal artery stenosis. *Stem Cells Transl Med* 14(4): szaf015.
11. Chen W, Lv L, Chen N, Cui E (2023) Immunogenicity of mesenchymal stromal/stem cells. *Scand J Immunol* 97(6): e13267.
12. Sanabria de la Torre R, Quiñones Vico MI, Fernández González A, Manuel Sánchez Díaz, Trinidad Montero Vilchez (2021) Alloreactive Immune Response Associated to Human Mesenchymal Stromal Cells Treatment: A Systematic Review. *J Clin Med* 10(13): 2991.
13. Hoseinzadeh A, Esmaeili SA, Sahebi R, Anahita Madani Melak, Mahmoud Mahmoudi, et al. (2025) Fate and long-lasting therapeutic effects of mesenchymal stromal/stem-like cells: mechanistic insights. *Stem Cell Res Ther* 16(1): 33.
14. Valencia J, Yáñez RM, Muntión S, María Fernández García, Jorge Diego MR et al. (2025) Improving the therapeutic profile of MSCs: Cytokine priming reduces donor-dependent heterogeneity and enhances their immunomodulatory capacity. *Front Immunol* 16: 1473788.
15. Huang XP, Sun Z, Miyagi Y (2010) Differentiation of allogeneic mesenchymal stem cells induces immunogenicity and limits their long-term benefits for myocardial repair. *Circulation* 122(23): 2419-2429.
16. Avivar Valderas A, Martín Martín C, Ramírez C, Borja Del Río, Ramón Menta, et al. (2019) Dissecting Allo-Sensitization After Local Administration of Human Allogeneic Adipose Mesenchymal Stem Cells in Perianal Fistulas of Crohn's Disease Patients. *Front Immunol* 10: 1244.
17. Song N, Scholtemeijer M, Shah K (2020) Mesenchymal Stem Cell Immunomodulation: Mechanisms and Therapeutic Potential. *Trends Pharmacol Sci* 41(9): 653-664.
18. Zhang J, Huang X, Wang H (2015) The challenges and promises of allogeneic mesenchymal stem cells for use as a cell-based therapy. *Stem Cell Res Ther* 6: 234.
19. Choi JR, Yong KW, Nam HY (2022) Current Status and Perspectives of Human Mesenchymal Stem Cell Therapy 2020. *Stem Cells Int* 2022: 9801358.
20. Eliopoulos N, Stagg J, Lejeune L, Pommey S, Galipeau J, et al. (2005) Allogeneic marrow stromal cells are immune rejected by MHC class I- and class II-mismatched recipient mice. *Blood* 106(13): 4057-4065.
21. Nauta AJ, Westerhuis G, Kruisselbrink AB, Lurvink EGA, Willemze R, et al. (2006) Donor-derived mesenchymal stem cells are immunogenic in an allogeneic host and stimulate donor graft rejection in a nonmyeloablative setting. *Blood* 108(6): 2114-2120.
22. Joswig AJ, Mitchell A, Cummings KJ (2017) Repeated intra-articular injection of allogeneic mesenchymal stem cells causes an adverse response compared to autologous cells in the equine model. *Stem Cell Res Ther* 8(1): 42.
23. Lohan P, Coleman CM, Murphy JM, Griffin MD, Ritter T, et al. (2014) Changes in immunological profile of allogeneic mesenchymal stem cells after differentiation: should we be concerned? *Stem Cell Res Ther* 5(4): 99.
24. Sun JM, Dawson G, Franz L (2020) Infusion of human umbilical cord tissue mesenchymal stromal cells in children with autism spectrum disorder. *Stem Cells Transl Med* 9(10): 1137-1146.
25. Bezstarosti S, Erpicum P, Maggipinto G, Geertje JD, Marlies EJ Reinders, et al. (2024) Allogeneic mesenchymal stromal cell therapy in kidney transplantation: should repeated human leukocyte antigen mismatches be avoided? *Front Genet* 15: 1436194.
26. Anwar IJ, Li S, Mulvihill M, Robin Schmitz, Brian Shaw. et al. (2025) Donor-specific mesenchymal cell infusion in human and nonhuman primate kidney transplantation. *Am J Transplant Off J Am Soc Transplant Am Soc Transl Surg* 25(10): 2114-2126.
27. Qu W, Yan D, Durand NC (2025) Intra-articular delivery of allogeneic bone marrow derived mesenchymal stromal cells (BM-MSCs) for painful lumbar facet arthropathy: a phase I clinical trial. *Stem Cell Res Ther* 16(1): 596.
28. Pang SHM, D Rozario J, Mendonca S, Tejasvini Bhuvan, Natalie L Payne, et al. (2021) Mesenchymal stromal cell apoptosis is required for their therapeutic function. *Nat Commun* 12(1): 6495.
29. Shokoohmand A, Patel NM, Braid L, Massimo Dominici, Tracy S P Heng, et al. (2026) Fate and function of exogenously administered mesenchymal stromal cells: current insights and future directions. *Cytotherapy* 28(2): 102007.
30. Rajendran RL, Gangadaran P (2026) Mesenchymal Stem Cells and Extracellular Vesicles: Bridging the Translational Gap in Regenerative Medicine. *Int J Mol Sci* 27(12): 5423.
31. (2024) U S Food and Drug Administration. FDA approves remestemcel-L-rknd for steroid-refractory acute graft versus host disease in pediatric patients.