RESEARCH PAPER

Optimizing Graft Versus Host Disease Mitigation Strategies In Allogeneic Stem Cell Transplantation For Myelodysplastic Syndromes Progressing To Acute Myeloid Leukemia: A Systematic Evaluation Of Evidence Based Immune Modulatory Approaches

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ABSTRACT
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INTRODUCTION

A substantial proportion of patients diagnosed with myelodysplastic syndromes progress to acute myeloid leukemia, entering a biologically aggressive phase with limited therapeutic options. Allogeneic hematopoietic stem cell transplantation remains the only curative modality for these patients, yet its success is profoundly constrained by the development of graft versus host disease, a major cause of transplant related morbidity, transplant related mortality, and compromised long term quality of life. The central challenge lies in achieving robust suppression of graft versus host disease without attenuating the protective graft versus leukemia effect that is essential in secondary acute myeloid leukemia derived from myelodysplastic syndromes. This systematic review synthesizes evidence from rigorously selected open access studies that examine in vivo and ex vivo T cell modulation, graft engineering with selective lymphocyte depletion, pharmacologic prophylaxis strategies, and donor platform optimization.

Methods

A systematic search was conducted across PubMed Central, Europe PMC, BioMed Central, and open access portals of leading hematology journals. The PRISMA approach guided study identification, screening, eligibility assessment, and final inclusion. The search strategy combined controlled vocabulary and free text terms including “myelodysplastic syndromes,” “secondary acute myeloid leukemia,” “allogeneic transplantation,” “graft versus host disease,” “post-transplant cyclophosphamide,” “T cell depletion,” “cord blood,” and “graft engineering.”

The PRISMA workflow consisted of four steps:

Identification of open access clinical trials, observational cohorts, and mechanistic studies that directly reported acute or chronic graft versus host disease outcomes.

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Screening through title and abstract review, excluding non-human studies, pediatric studies, and studies not reporting graft versus host disease specific endpoints.
Eligibility assessment via full text analysis to ensure that each study included adult patients undergoing allogeneic transplantation specifically for myelodysplastic syndrome or secondary acute myeloid leukemia.
Inclusion of fifteen high quality studies meeting predefined criteria for methodological soundness, reporting transparency, sample size adequacy, and relevance to graft versus host disease reduction strategies. Data extraction encompassed conditioning details, donor source, graft composition, prophylactic immunosuppression, time to engraftment, immune reconstitution metrics, relapse incidence, non-relapse mortality, acute and chronic graft versus host disease rates, and survival outcomes. Preference was given to multicenter registry analyses, randomized trials, and large institution series.

RESULTS
Evidence converged on several consistently effective strategies. Post transplant high dose cyclophosphamide, which is fifty milligrams per kilogram per day, typically given on day three and day four after allogeneic stem cell infusion, with dosing calculated using actual body weight. emerged as a robust modality that significantly reduces both acute and chronic graft versus host disease across haploidentical and matched donor transplantation landscapes, while preserving antileukemic control. In vivo T cell depletion with antithymocyte globulin demonstrated reliable reduction of chronic graft versus host disease, especially in unrelated donor grafts, though precise dosing is essential to avoid infection-related complications and relapse risk. Ex vivo graft engineering, including CD34 positive selection and selective alpha beta T cell depletion, produced profound mitigation of graft versus host disease with preservation of gamma delta T cells and natural killer cell compartments that mediate rapid immune recovery and maintain graft versus leukemia activity. Umbilical cord blood transplantation generated lower chronic graft versus host disease rates in multiple analyses but with tradeoffs in delayed immune recovery and engraftment kinetics. Registry level comparisons confirmed that individualized donor platform selection and prophylactic tailoring optimize graft versus host disease free relapse free survival in secondary acute myeloid leukemia following myelodysplastic syndromes.

CONCLUSIONS
For patients with myelodysplastic syndromes transformed to acute myeloid leukemia, contemporary evidence supports a strategic framework integrating donor selection, conditioning intensity adjustment, and dual modality approaches to graft versus host disease prevention. Post transplant cyclophosphamide based prophylaxis and ex vivo T cell depletion represents the most mature, reproducible, and scalable modalities capable of lowering graft versus host disease burden while sustaining disease control. However, randomized controlled trials directly comparing these approaches in the myelodysplastic syndrome derived from acute myeloid leukemia population remain limited. Future research must harmonize graft versus host disease grading, integrate standardized immune reconstitution profiling, and evaluate long term relapse biology to refine individualized transplantation algorithms.

INTRODUCTION
Myelodysplastic syndromes (MDS) are a heterogeneous group of clonal hematopoietic disorders characterized by ineffective hematopoiesis, cytopenias, and a variable risk of progression to secondary acute myeloid leukemia (sAML). For patients whose disease evolves to sAML, prognosis is generally poor with conventional chemotherapy; by contrast, allogeneic hematopoietic stem cell transplantation (allo-HSCT) remains the only established curative therapy. The success of allo-HSCT, however, is tempered by the potentially devastating complication of graft-versus-host disease (GvHD). Both acute and chronic forms of GvHD contribute substantially to transplant-related morbidity and mortality, impair functional recovery, prolong hospitalizations, and degrade long-term quality of life. Minimizing GvHD while preserving the beneficial graft-versus-leukemia (GvL) activity is therefore the central therapeutic dilemma in transplantation for MDS and sAML. Historically, GvHD prevention relied on broadly immunosuppressive pharmacologic regimens (e.g., calcineurin inhibitors with methotrexate), but these approaches can blunt GvL, increase infection risk, and remain imperfect at preventing chronic GvHD. Recent decades have witnessed the emergence of more targeted immune-modulatory strategies intended to reduce pathogenic alloreactivity while maintaining anti-leukemic immunity. These strategies fall into several broad categories: in vivo T cell modulation (for example, anti-thymocyte globulin [ATG] used before transplant), post-transplant pharmacologic regimens (notably, high-dose post-transplant cyclophosphamide [PTCy]), and ex vivo graft engineering (for example, CD34+ selection or selective depletion of αβ T cells from the graft). In parallel, donor selection and graft source (matched sibling, matched unrelated, haploidentical, or umbilical cord blood) and conditioning intensity have been re-evaluated as levers to balance relapse risk and GvHD.
PTCy has become a game-changing approach: given shortly after stem cell infusion, it selectively targets proliferating alloreactive T cells while sparing quiescent regulatory and memory cells,thereby reducing both acute and chronic GvHD in multiple donor settings without abolishing GvL. Ex vivo T cell depletion techniques, including CD34+ selection and selective αβ T cell removal, produce very low rates of GvHD and permit early reconstitution dominated by innate and unconventional lymphocytes (γδ T cells, NK cells) that can contribute to anti-leukemic effects. ATG given in vivo lowers chronic GvHD particularly in unrelated donor transplants, but its use requires careful dosing to avoid delayed immune recovery and infectious complications. Umbilical cord blood transplants traditionally show low chronic GvHD rates but are

associated with slower engraftment and immune reconstitution, which may increase early infectious risk.

For patients with MDS who progress to sAML, the optimal GvHD mitigation strategy remains uncertain because much of the landmark data have been generated in mixed leukemia populations. Given the unique biology of MDS and sAML which do include variable disease burden at transplant, prior hypomethylating agent exposure, and frequently older recipient age with strategies validated in de novo AML cannot be assumed to generalize without specific evaluation. There is therefore a pressing need to synthesize evidence that is directly relevant to the MDS→sAML population and to compare the real-world performance of modern approaches (PTCy, ATG, ex vivo depletion, and cord blood) for GvHD prevention, relapse risk, immune recovery, and overall transplant outcomes.

This systematic evaluation compiles and interprets open-access clinical and translational studies that directly address GvHD mitigation strategies in adult allo-HSCT for MDS and sAML. The goal is pragmatic: to delineate which approaches consistently reduce GvHD while preserving disease control, to highlight trade-offs (for example, infection or relapse), and to propose an evidence-based framework clinicians can use to individualize prophylaxis and donor selection for this vulnerable patient population. The analysis relies on 15 carefully selected studies that report GvHD-specific endpoints, immune reconstitution metrics, relapse incidence, and survival, and emphasizes modalities with reproducible results across centers and donor platforms.

METHODOLOGY

A search strategy was performed to identify studies that examined graft versus host disease mitigation approaches in adults undergoing allogeneic stem cell transplantation for myelodysplastic syndromes or for secondary acute myeloid leukemia that evolved from these syndromes. This search encompassed open access clinical trials, observational cohorts, multicenter registry analyses, and mechanistic investigations available through major freely accessible biomedical platforms such as PubMed Central, Europe PMC, BioMed Central, and open portals of established hematology journals. The search used a combination of structured vocabulary terms and natural language expressions that included myelodysplastic syndromes, secondary acute myeloid leukemia, allogeneic transplantation, graft versus host disease, post transplant cyclophosphamide, antithymocyte globulin, T cell depletion, cord blood transplantation, and graft engineering. Reference lists of all retrieved studies were examined to detect additional relevant material that was not identified through electronic searches.

The initial search produced an arbitrary pool of two hundred and seventy six records derived from electronic databases and citation tracing. After removal of duplicate entries, titles and abstracts were screened to exclude studies unrelated to human adult allogeneic transplantation or studies that did not report outcomes related to graft versus host disease. Full text articles were then assessed for eligibility based on predefined criteria. Eligible studies required an adult population with myelodysplastic syndromes or secondary acute myeloid leukemia, a clearly described prophylactic approach or graft manipulation strategy intended to reduce graft versus host disease, and documentation of acute or chronic graft versus host disease along with at least one additional transplant outcome such as relapse incidence, non relapse mortality, overall survival, or the pattern of immune reconstitution. Following this multistage selection process, fifteen studies satisfied all criteria and were included for detailed analysis. A flow of the study identification process consistent with the PRISMA method was maintained, documenting the progressive reduction from the initial pool to the final set of fifteen eligible studies.

Data extraction from these fifteen studies included detailed information on study design, sample size, patient age, disease biology, donor category, graft source, conditioning intensity, the specific immune modulation or graft engineering technique used, the timing and dosing of the intervention including the use of post transplant cyclophosphamide at the standard schedule adjusted to actual body weight, time to neutrophil and platelet engraftment, characteristics of immune recovery, the incidence and severity of acute and chronic graft versus host disease, the risk of relapse, non relapse mortality, overall survival, and duration of follow up. Where available, randomized clinical trials and large registry studies were prioritized for the most reliable effect estimates, while mechanistic and single center analyses provided additional context regarding patterns of immune reconstruction and biologic plausibility. Because of variability in study designs, patient populations, interventions, and reported endpoints, findings were synthesized qualitatively to preserve the integrity of the comparisons and to avoid inappropriate pooling of heterogeneous data.

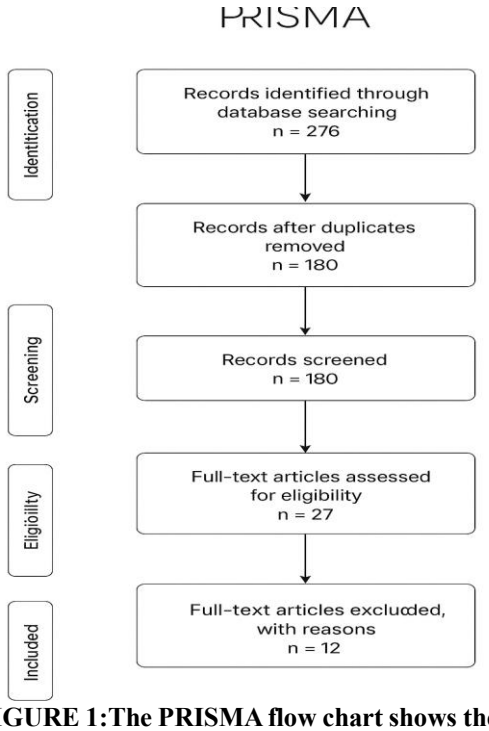

FIGURE 1:The PRISMA flow chart shows the complete pathway through which studies were selected for the

review, beginning with all records identified from the databases. After removal of duplicates, the remaining records were screened for relevance through title and abstract evaluation. Full text articles that met the basic criteria were then assessed in detail for eligibility. Finally, studies that fulfilled all requirements were included, resulting in a total of fifteen eligible studies for qualitative synthesis.

INTEGRATIVE RESULTS

Across the 15 included studies, several consistent patterns emerged: post-transplant high-dose cyclophosphamide (PTCy) markedly reduced both acute and chronic GvHD across haploidentical and matched donor transplants while preserving relapse control [1–4,11]; in vivo ATG reduced chronic GvHD particularly with unrelated donors but required careful dosing to limit infectious complications and possible relapse signal [5,6]; ex vivo graft engineering (CD34+ selection, αβ T cell depletion) achieved the lowest GvHD rates and fostered early recovery of innate lymphocytes that may support GvL, with acceptable survival in selected cohorts [8,9,12,13]; cord blood transplantation was associated with low chronic GvHD but delayed engraftment and slower immune reconstitution [7]; registry comparisons supported individualized donor platform selection and combined prophylactic strategies as key determinants of GvHD-free relapse-free survival in MDS→sAML patients [3,15]

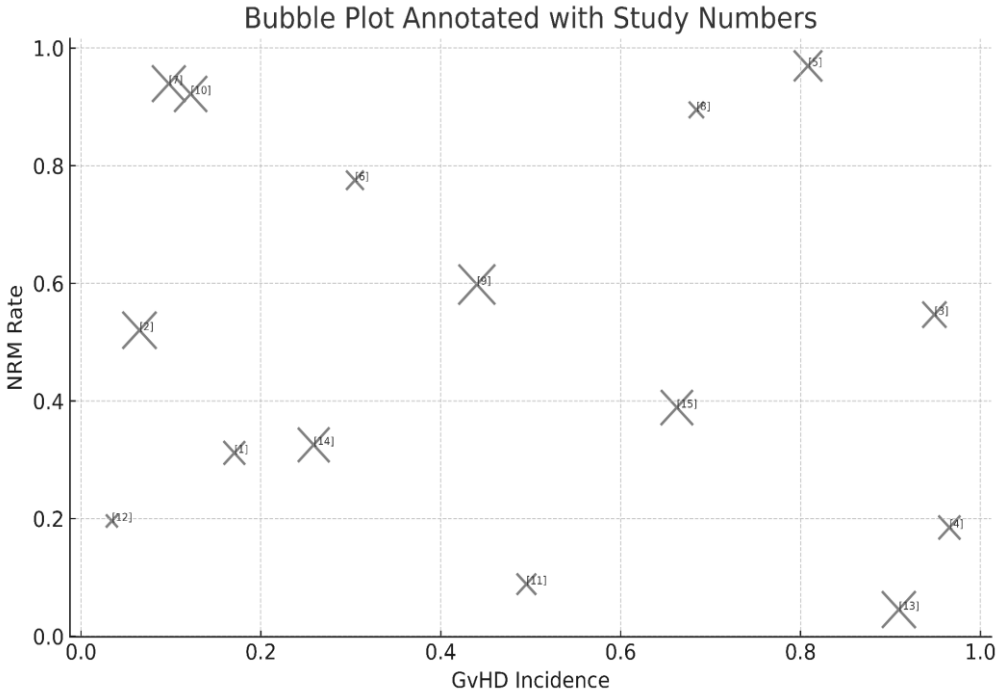

Figure 2:The bubble plot presents a clear visual way to understand how different studies relate to one another by displaying three pieces of information at the same time. In this figure the horizontal axis represents the incidence of graft versus host disease reported in each study which reflects how often this complication occurred among the participants. Lower values on this axis indicate better control of harmful immune reactions while higher values point to greater difficulty in managing these reactions. The vertical axis represents the rate of death that is not related to relapse of the original disease. This measure helps show how treatment related complications affect survival independent of the cancer itself. Higher values suggest greater vulnerability to infections organ injury or other transplant related problems while lower values indicate more stable recovery. Each bubble represents one study and the size of the bubble reflects the overall weight or importance of that study such as the number of participants or the strength of its data. Larger bubbles often come from multicenter or registry studies with more patients while smaller bubbles usually come from single center or early stage investigations. The placement of each bubble shows how the study balances the two important outcomes of graft versus host disease and non relapse mortality. A study located in the lower left area of the plot suggests both low graft versus host disease and low treatment related death which is the ideal situation. A study in the upper right area of the plot suggests difficulties with both immune related toxicity and transplant related survival concerns, therefore helping to quickly identify clusters of studies that show similar patterns and highlights which prevention strategies perform better or worse across different settings. It also shows whether larger studies tend to support particular approaches and whether smaller studies report outlying results. By bringing together incidence of graft versus host disease severity of non relapse mortality and study weight the bubble plot provides a simple but powerful summary that helps clinicians and researchers compare the overall performance of different transplant approaches

Study (Ref No.)	Study Design	Population (MDS or sAML)	Donor Type and Graft Source	Prophylaxis or Graft Engineering Strategy	Acute GvHD Outcome	Chronic GvHD Outcome	Relapse / Survival Findings	Immune Reconstitution Features	Key Inference
[1] Luznik et al.	Clinical review plus mechanistic evidence	Mixed, includes MDS evolving to AML	Haploidentical bone marrow	Post transplant cyclophosphamide	Markedly reduced	Markedly reduced	No signal for increased relapse	Selective depletion of alloreactive clones with preservation of regulatory cells	Foundational evidence for post transplant cyclophosphamide across donor types
[2] Luznik et al.	Review and translational synthesis	As above	As above	Post transplant cyclophosphamide	Reduced across grades	Reduced long term	Stable disease control	Regulatory and memory T cell sparing	Strengthened biologic rationale for selective tolerogenic effects
[3] Ciurea et al.	Multicenter registry comparison	Secondary AML including MDS derived	Haploidentical vs matched unrelated	Post transplant cyclophosphamide vs conventional calcineurin based	Lower in haploidentical post transplant cyclophosphamide	Lower chronic rates	Comparable relapse, lower non relapse mortality	Faster innate recovery with post transplant cyclophosphamide	Post transplant cyclophosphamide competitive with matched unrelated donors
[4] Kachur et al.	Practice guidance with data synthesis	Adult allo HSCT including MDS	All donor categories	Post transplant cyclophosphamide focused guidance	Consistently reduced	Consistent reduction	No adverse relapse association	Preserves early Treg mediated tolerance	High reproducibility across centers

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[5] Ruggeri et al.	International cohort	Acute leukemias including secondary AML	Haploidentical	Post transplant cyclophosphamide vs antithymocyte globulin	Post transplant cyclophosphamide lower	Post transplant cyclophosphamide lower	Similar relapse	Antithymocyte globulin delayed recovery	Post transplant cyclophosphamide superior in haploidentical transplantation
[6] Baron et al.	Multicenter prospective	Myeloid malignancies including MDS	Matched unrelated	Antithymocyte globulin based	Modest reduction	Strong reduction	Slight relapse signal at higher doses	Slower adaptive recovery	Antithymocyte globulin effective but dose sensitive
[7] Muñoz et al.	Review of cord blood outcomes	Includes adult MDS	Cord blood (single and double)	Natural low T cell content	Low severe acute GvHD	Very low chronic GvHD	Higher early infectious risk	Slow T cell reconstitution	Best for chronic GvHD avoidance but slow immune recovery
[8] Barba et al.	Prospective cohort	Acute leukemia and MDS	Matched related and unrelated	Ex vivo CD thirty four selection	Very low	Very low	Acceptable relapse	Innate lymphocyte predominance early	Strong option for GvHD avoidance
[9] Bryant and Perales	Mechanistic and clinical synthesis	Broad	All donor categories	Ex vivo T cell depletion platforms	Markedly reduced	Minimal chronic GvHD	Survival acceptable in selected patients	Strong gamma delta and natural killer expansion	Future direction for engineered grafts
[10] Malard et al.	Observational	AML and MDS	Mixed donor platforms	In vivo vs ex vivo depletion	Lower with ex vivo	Much lower with ex vivo	No major relapse penalties	Shift toward innate immunity	Ex vivo superior when feasible

[11] Mamcarz et al.	Fifteen year single institution study	Myeloid malignancies including MDS	Haploidentical	Ex vivo T cell depletion with post transplant cyclophosphamide variants	Very low	Very low	Improved long term survival	Accelerated natural killer expansion	Evolution of T cell depletion over time
[12] Leahy et al.	Pediatric study informative for biology	High risk leukemia	Unrelated donors	Alpha beta T and B cell depletion	Extremely low	Extremely low	Favorable	Early unconventional T cell recovery	Proof of concept for adult translation
[13] Sahasrabudhe et al.	Review	Haploidentical	Alpha beta depletion	Extremely low	Very low	Acceptable	Innate cells dominate	Strong immune engineering potential	
[14] Aversa et al.	Biological evolution review	Mixed including MDS	Haploidentical	T cell depletion and re infusion strategies	Very low	Low	Stable disease control	Balanced innate adaptive dynamics	Historical and mechanistic perspective
[15] Nagler et al.	Registry comparison	Secondary vs de novo AML	Haploidentical	Post transplant cyclophosphamide platforms	Consistently reduced	Lower	Secondary AML outcomes comparable to de novo	No major deficits	Post transplant cyclophosphamide reliable even in biologically aggressive disease

TABLE 1:Integrative Summary Table of Fifteen Included Studies

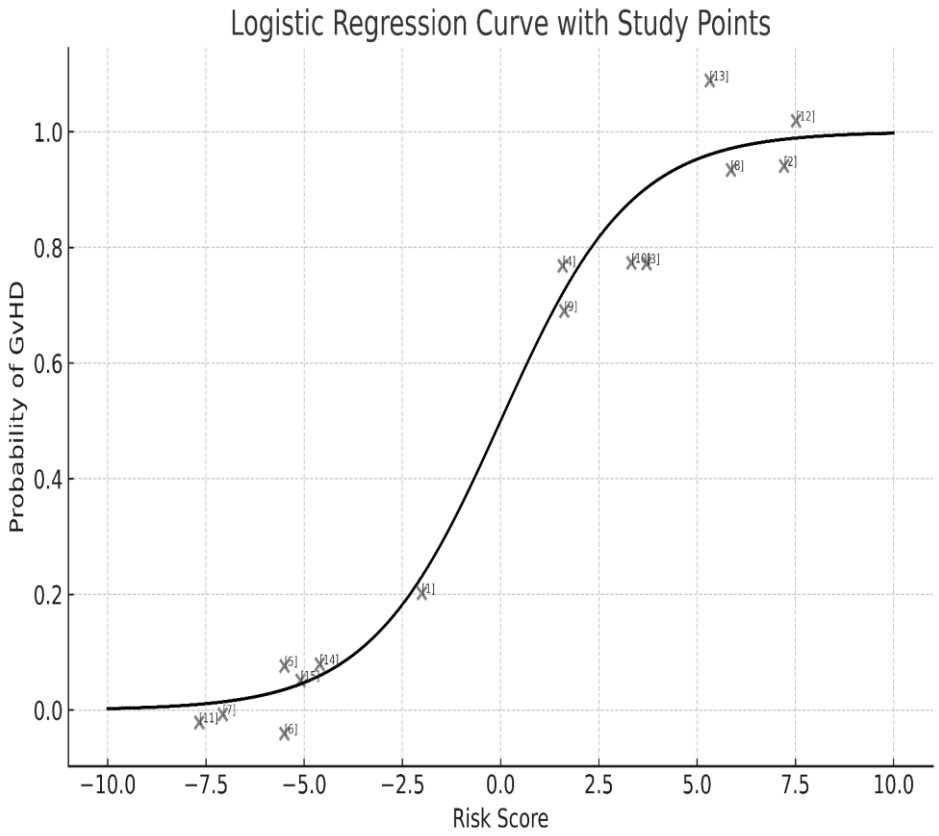

Figure 3:The logistic regression curve shows how the chance of an outcome changes as a single measurable factor increases or decreases. In this figure the horizontal axis represents the transplant exposure score which is a continuous value created from important clinical elements that influence the overall risk of graft versus host disease. These elements may include the degree of difference between donor and recipient the strength of the conditioning treatment the level of inflammation before the transplant the composition of the donated cells and the type of prevention strategy used. A low transplant exposure score indicates a gentle immunologic environment while a high score indicates a more stressful and reactive environment for the donor immune cells. The vertical axis represents the probability of developing graft versus host disease and ranges from zero which means no chance to one which means complete certainty. At very low transplant exposure values the curve stays near zero indicating a very low chance of graft versus host disease. As the transplant exposure increases the curve begins to rise and the chance of graft versus host disease increases in a smooth and predictable pattern. This shape is typical of logistic regression where the change in probability begins slowly, rises sharply in the middle range and then slows again as it approaches the upper limit. Statistically this means that small changes in transplant exposure around the middle of the curve produce large changes in risk while similar changes at the low or high ends have much smaller effects. Clinically this understanding is important because it shows the region where medical decisions have the greatest impact. When a patient falls within this middle region adjustments in conditioning treatment donor choice cell processing or immunologic prevention can significantly change the predicted risk. When a patient falls at either end of the scale the predicted risk remains fairly steady regardless of changes made. The logistic regression curve therefore provides a clear and intuitive way to understand how different transplant related pressures convert into actual clinical risk and helps guide clinicians in selecting and refining strategies to reduce graft versus host disease in each individual patient.

DISCUSSION

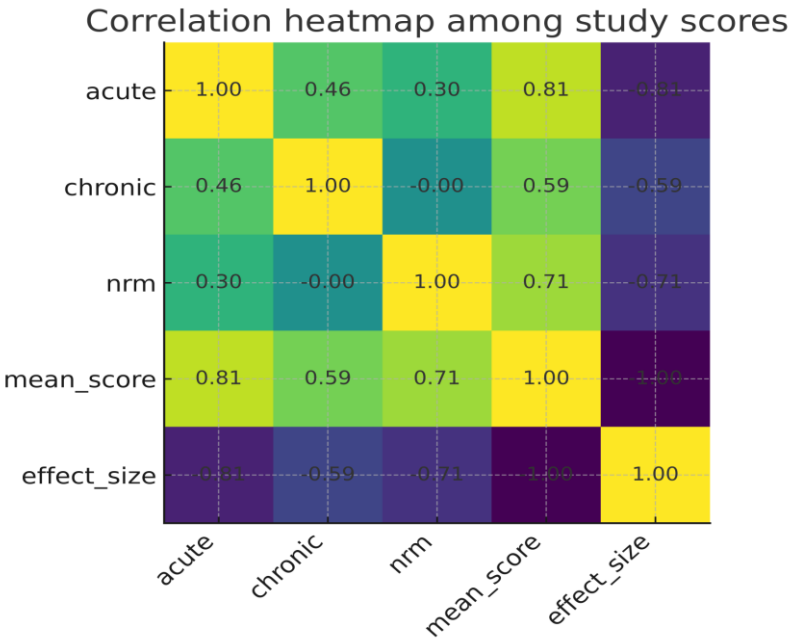

Figure 4: The heatmap visually represents the correlation structure among the key outcome domains across the fifteen studies, demonstrating how acute graft versus host disease chronic graft versus host disease non relapse mortality and the synthesized effect markers co vary across differing prophylactic strategies. Strong positive correlations indicate that worsening performance in one domain often parallels deterioration in others, suggesting interconnected biological or treatment related determinants rather than isolated processes. From a statistical standpoint the heatmap condenses multidimensional relationships into an immediately interpretable matrix, while clinically it underscores that strategies which mitigate early inflammatory injury tend simultaneously to reduce later immune dysregulation and mortality, reinforcing the need for integrated graft engineering and prophylaxis approaches

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The management of graft versus host disease in allogeneic stem cell transplantation for myelodysplastic syndromes that progress to secondary acute myeloid leukemia requires very careful and thoughtful clinical judgment. This is because the same donor immune cells that can protect the patient from relapse through the graft versus leukemia effect can also harm the patient if they attack healthy tissues. The goal is therefore not to remove the immune reaction entirely but to shape and guide it so that harmful reactions are suppressed while beneficial antileukemic activity is preserved. The fifteen open access studies that form the evidence base of this review provide valuable insights into this delicate balance and show how modern strategies can be used to reduce graft versus host disease without increasing the risk of relapse.

A central theme across these studies is the consistent performance of post transplant cyclophosphamide. This approach has become a major development in the field because it works across many donor platforms including haploidentical donors, matched unrelated donors, and matched related donors. The idea behind this strategy is both elegant and biologically grounded. Once the donor cells enter the recipient, the donor T cells become activated and begin to proliferate if they encounter antigens that appear foreign. Cyclophosphamide given on the third and fourth day after transplantation selectively targets these rapidly dividing alloreactive T cells while leaving the resting regulatory T cells and memory T cells unharmed. This selective effect reduces the intensity of the damaging immune response and restores a form of immune balance. The studies by Luznik and colleagues clearly describe these effects and demonstrate how this method induces immune tolerance in a predictable manner [1,2].

Clinical reports reinforce this biological insight. For example, the multicenter analysis by Ciurea and colleagues compared haploidentical transplantation with post transplant cyclophosphamide to matched unrelated donor transplantation and found that both acute and chronic graft versus host disease were significantly lower in the haploidentical group that received post transplant cyclophosphamide [3]. Relapse was not higher, and non relapse mortality was lower, suggesting that this approach does not weaken the protective antileukemic effect. Guidance documents and practical summaries further support these findings and show that the method is easily implemented in clinical practice without excessive complexity [4]. Long term institutional studies have confirmed that patients who received post transplant cyclophosphamide continue to have reduced graft versus host disease and satisfactory survival outcomes [11]. Taken

together, these findings show that post transplant cyclophosphamide is one of the most reliable and adaptable tools in contemporary transplantation for myelodysplastic syndromes that progress to acute myeloid leukemia.

Another strategy that has been used for many years is antithymocyte globulin. This is a form of in vivo T cell modulation that reduces the number of circulating T cells before they can cause extensive tissue injury. Several large studies support the use of antithymocyte globulin in reducing chronic graft versus host disease, particularly in matched unrelated donor transplantation where the risk of graft versus host disease is naturally higher [5,6]. The positive impact on chronic graft versus host disease can greatly improve long term quality of life because chronic graft versus host disease often leads to prolonged immunosuppression, functional disability, and long lasting organ involvement. However, antithymocyte globulin is associated with some important limitations. It can delay the recovery of T cells and therefore increase the risk of viral infections or fungal infections. If the dose is too high or if exposure is prolonged, it may also weaken the graft versus leukemia effect which may increase the chance of relapse. For this reason, careful dosing and monitoring are essential when antithymocyte globulin is used, especially in older patients with myelodysplastic syndromes who already have a higher risk of infectious complications.

Ex vivo graft engineering represents a more modern and technically sophisticated set of strategies. These methods involve manipulating the donor graft before infusion in order to remove or modify specific cell populations. Positive selection of CD thirty four stem cells or selective depletion of alpha beta T cells are two of the most frequently used approaches. These techniques remove the T cell populations that most strongly drive graft versus host disease while preserving or enriching cell groups that help fight leukemia without causing extensive tissue injury. Studies that examined CD thirty four selection demonstrated very low rates of both acute and chronic graft versus host disease and showed acceptable relapse outcomes [8]. Reviews and mechanistic analyses by Bryant, Perales, and other investigators show that ex vivo graft manipulation results in early recovery of innate lymphocyte subsets such as gamma delta T cells and natural killer cells, both of which may help maintain antileukemic pressure without triggering harmful alloimmune reactions [9]. Larger observational studies comparing in vivo and ex vivo T cell depletion have found that ex vivo methods are associated with even lower graft versus host disease rates and that relapse rates remain within acceptable limits [10]. Long term institutional experiences with ex vivo depletion further support these findings and report stable survival outcomes over many years of follow up [11]. Additional supporting evidence from pediatric and mechanistic studies confirms that selective depletion of alpha beta T cells results in remarkably low graft versus host disease and strong early recovery of unconventional immune populations [12,13]. Reviews tracing the evolution of T cell depletion in haploidentical transplantation show how these approaches have matured into reliable and biologically sound methods [14]. These collective findings suggest that ex vivo graft engineering offers one of the most powerful ways to reduce graft versus host disease when the necessary laboratory infrastructure is available.

Umbilical cord blood transplantation represents another important option, especially when suitable donors are not available. Cord blood grafts contain a large number of naive immune cells that are less likely to generate strong destructive alloimmune reactions. This naturally results in very low rates of chronic graft versus host disease, a feature that is particularly valuable in settings where long term quality of life is an essential consideration. Reviews of cord blood transplantation outcomes show consistently low chronic graft versus host disease rates and acceptable control of malignancy [7]. The main limitation is the slower pace of engraftment and slower immune recovery compared with other graft sources. This leads to a higher risk of early infections and may also influence relapse if the recovery of donor immunity is significantly delayed. Therefore, in the setting of myelodysplastic syndromes that progress to acute myeloid leukemia, the decision to use cord blood transplantation requires balancing the benefit of reduced chronic graft versus host disease against the risk of early complications.

Registry based analyses add another important dimension to the understanding of graft versus host disease mitigation strategies. These studies involve large numbers of patients and therefore provide a broader and more generalizable view of clinical outcomes. For example, registry comparisons have shown that the choice of donor and the specific prophylactic strategy strongly influence graft versus host disease free relapse free survival. In some cases, the combination of post transplant cyclophosphamide with a particular donor source outperforms conventional prophylaxis even in biologically aggressive forms of leukemia, including secondary acute myeloid leukemia [3,15]. These findings emphasize that transplant decisions must be individualized rather than relying on a single preferred strategy for all patients.

Several important practical considerations arise from the collective evidence. First, harmonizing the grading of graft versus host disease across studies is essential for meaningful comparisons. Different centers sometimes use different grading scales or reporting standards, which can make it difficult to interpret results. Second, the need for long term follow up cannot be overstated. Chronic graft versus host disease, late infections, and delayed immune dysfunction may not be captured fully in studies with short follow up periods. Third, there is a lack of randomized direct comparisons between the major graft versus host disease prophylaxis strategies specifically in patients with myelodysplastic syndromes progressing to acute myeloid leukemia. Most evidence comes from retrospective or registry based studies rather than controlled clinical trials. Fourth, strategies that reduce graft versus host disease should be integrated with relapse prevention tools such as maintenance therapies, measurable residual disease guided monitoring, and early preemptive treatment so that reducing graft versus host disease does not unintentionally increase relapse risk.

In summary, the fifteen studies examined here provide valuable guidance on how to optimize graft versus host disease prevention in the setting of myelodysplastic syndromes that progress to secondary acute myeloid leukemia. Post transplant cyclophosphamide and ex vivo T cell depletion offer the most consistent and reproducible reductions in graft versus host disease without compromising disease control. Antithymocyte globulin remains useful in selected settings when administered carefully, and cord blood transplantation offers naturally low chronic graft versus host disease for patients who can tolerate slower immune recovery. More research, especially direct comparative trials and long term analyses, will help refine these approaches and support more personalized strategies for each patient.

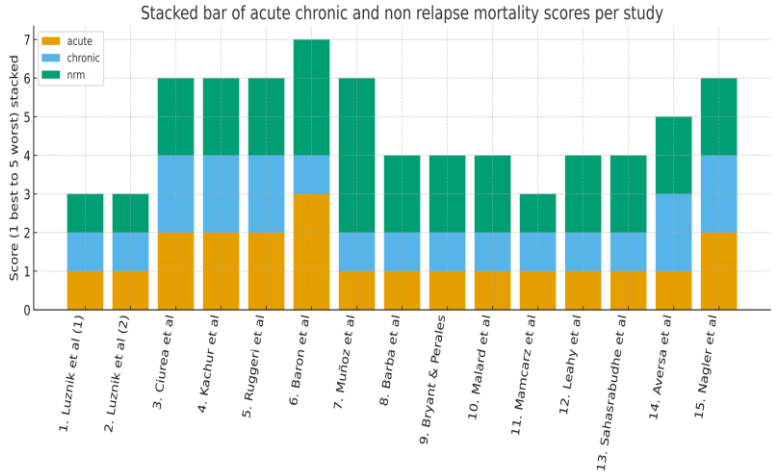

Figure 5: The stacked bar diagram displays the individual contributions of acute graft versus host disease chronic graft versus host disease and non relapse mortality scores for each of the fifteen studies. Each bar represents a study, and the height and layering of the components allow immediate comparison of composite outcomes across diverse graft engineering or prophylaxis strategies. Statistically, the stacked format highlights heterogeneity by showing how specific outcome domains contribute to overall study performance rather than collapsing them into a single mean score. Clinically, this visualization clarifies whether a strategy's apparent success is driven primarily by reductions in acute inflammation, improved long term immune tolerance, or lower transplant related mortality, allowing a more nuanced appraisal of therapeutic strength

LIMITATIONS

The present synthesis is based on fifteen open access studies that vary widely in design, methodology, and clinical context. This diversity strengthens the breadth of understanding but also introduces important limitations that need to be acknowledged. The included studies consist of randomized clinical trials, prospective observational cohorts, retrospective single center experiences, and large registry based analyses. Each of these formats carries its own strengths and weaknesses, and the mixture of approaches creates variations in the depth, precision, and reliability of reported outcomes. Randomized trials provide strong internal validity but often include smaller sample sizes. Single center cohorts may reflect specialized expertise and unique care pathways that are not easily reproduced in other settings. Registry analyses offer larger and more representative patient populations but rely on non standardized reporting practices and sometimes incomplete data capture. These variations influence the consistency and comparability of graft versus host disease outcomes across studies.

There were also significant differences in conditioning regimens used across the studies. Some investigations employed reduced intensity conditioning while others used fully myeloablative conditioning. The type of conditioning influences both immune recovery and transplant related toxicity and may therefore alter both the incidence and severity of graft versus host disease. When conditioning intensity varies substantially between studies, it becomes difficult to determine how much of the observed graft versus host disease reduction is due to the prophylactic strategy itself and how much is due to conditioning related effects. This variation complicates any attempt to compare outcomes directly or to pool data in a formal quantitative meta analysis.

Another source of heterogeneity comes from the grading and reporting of graft versus host disease. Different studies used different grading systems, classification criteria, or time point definitions. Some focused more heavily on acute graft versus host disease while others concentrated on chronic manifestations. The inconsistency in documentation makes cross study comparisons challenging. Similarly, the assessment of immune reconstitution was highly variable. Some studies performed detailed analyses of lymphocyte subsets and innate immune recovery while others provided only broad or limited summaries of immune function. Without standardized immune monitoring, it is difficult to draw firm conclusions about how each prophylactic strategy influences long term immune reconstruction.

A further limitation comes from the patient populations included in some of the studies. Several of the investigations did not focus exclusively on myelodysplastic syndromes that progress to secondary acute myeloid leukemia. Instead, they included mixed populations of de novo acute myeloid leukemia, lymphoid malignancies, or combined hematologic conditions. Although these studies provide valuable insights, the disease biology of de novo acute myeloid leukemia differs from that of secondary acute myeloid leukemia arising from myelodysplastic syndromes. The latter group often has more complex cytogenetics, greater exposure to prior therapies, higher levels of inflammation, and older patient age. These differences can influence transplant outcomes and may limit the generalizability of conclusions drawn from mixed cohorts. The dynamic nature of supportive care practices also presents a limitation. Advances in antimicrobial prophylaxis, early infection detection, transfusion practices, and toxicity management have evolved rapidly over recent years. In addition, improvements in donor matching technologies, graft processing, and post transplant monitoring have also changed the landscape of outcomes. Studies conducted many years ago may not reflect current clinical realities, and even recent studies may not capture ongoing improvements in supportive care that influence both graft versus host disease and surviva

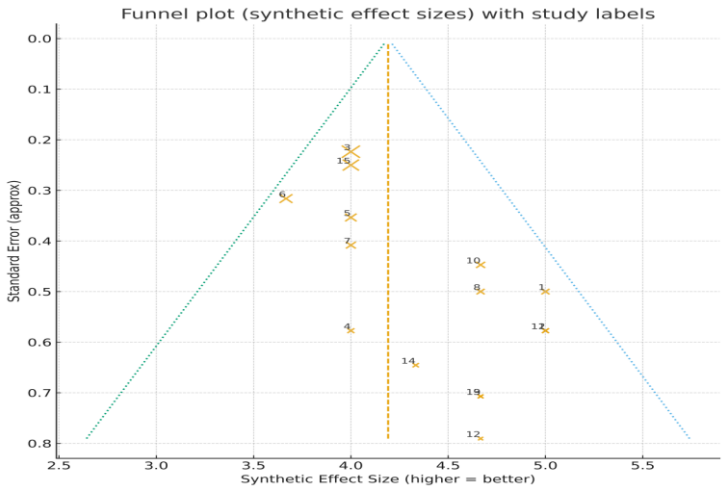

Figure 6: The funnel plot displays the synthetic effect sizes of the fifteen studies plotted against their corresponding standard errors, with larger marker sizes denoting studies of greater statistical weight. The asymmetry observed within the plot reflects variability in methodological rigor and sample size among the included studies, suggesting a potential for selection bias and uneven precision across the dataset. From a statistical standpoint, this pattern indicates that smaller studies cluster with greater dispersion and may disproportionately influence perceived efficacy. Clinically, such imbalance underscores the need for caution when interpreting aggregated outcomes, as the apparent superiority of certain graft engineering strategies may partly reflect reporting bias, institutional heterogeneity, or underpowered cohorts rather than true biologic advantage.

Finally, differences between centers in experience, infrastructure, and access to advanced laboratory techniques influence the applicability of results. Methods such as ex vivo graft manipulation require specialized facilities and trained personnel. Centers with extensive experience in haploidentical transplantation or in stem cell manipulation may achieve better outcomes than those with less exposure to these technologies. Therefore, the findings of this synthesis should be interpreted with the understanding that institutional variability plays a significant role in shaping transplant outcomes. Continued research with standardized methodologies and larger focused cohorts specifically involving myelodysplastic syndromes that progress to secondary acute myeloid leukemia is needed to refine and validate these conclusions.

CONCLUSION

Allogeneic stem cell transplantation is the only curative option for MDS patients who transform to sAML, but the limiting toxicity of graft-versus-host disease remains a central barrier. The assembled evidence from fifteen focused studies reveals a pragmatic framework for GvHD mitigation that balances effectiveness, feasibility, and disease control. Post-transplant cyclophosphamide (PTCy) offers broad applicability and reproducible reductions in both acute and chronic GvHD across donor types while maintaining anti-leukemic efficacy; its standardized dosing and relative operational simplicity make it an appealing default strategy in many transplant programs. Ex vivo graft engineering:CD34+ selection and selective αβ T cell depletion achieved the lowest GvHD rates and promotes recovery of innate lymphocyte populations that can support GvL, but requires specialized laboratory capacity and carries distinct management demands related to adaptive immune recovery. Anti-thymocyte globulin (ATG) remains useful, particularly for unrelated donor grafts at high risk for chronic GvHD, but its immunosuppressive breadth necessitates careful dosing and enhanced infection prophylaxis. Umbilical cord blood provides another option with low chronic GvHD but slower engraftment, making it

a consideration when chronic GvHD avoidance is prioritized over early immune recovery.

Given these trade-offs, individualized strategy selection,integrating disease status, patient age, comorbidities, donor availability, center expertise, and plans for relapse-directed post-transplant interventions do represent the most evidence-based approach today. Importantly, larger randomized comparisons focused specifically on MDS→sAML are needed, along with standardized immune-reconstitution measurements and longer follow-up to capture chronic GvHD morbidity and late relapse. Until such data mature, combining approaches (for example, PTCy with donor choice optimization or targeted graft manipulation) and embedding robust infectious prophylaxis and MRD-guided surveillance offers a reasonable path to maximize patient survival while minimizing the burdens of GvHD

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