AML - 1455

IMMUNE-CYTOKINE SIGNATURE AND VITAMIN D ASSOCIATED WITH CLINICAL REMISSION STATUS IN ACUTE MYELOID LEUKEMIA

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INTRODUCTION

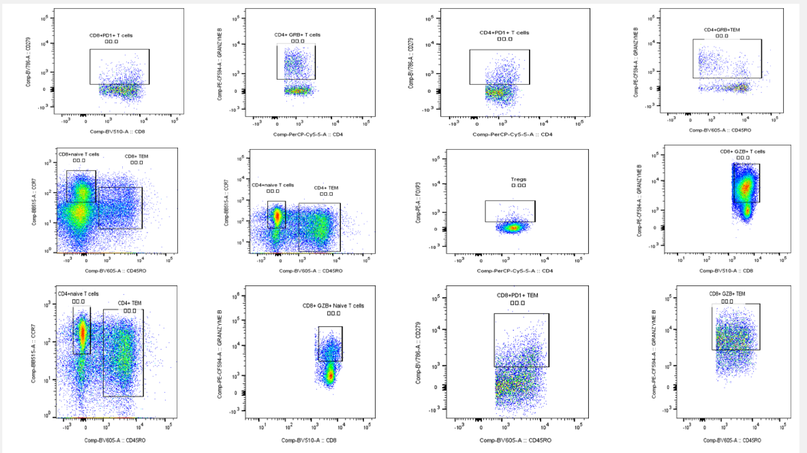
Immune dysfunction is a hallmark of acute myeloid leukemia (AML), affecting disease progression and treatment outcomes. Vitamin D, a lesser-known immunomodulator, may influence immune cell differentiation and effector functions. However, integrative analyses assessing the combined effects of vitamin D status and immune profiles on clinical remission in AML remain limited.

AIM

To investigate the relationship between immune phenotypes, Vitamin D levels, and clinical outcomes in AML patients using multicolor flow cytometry and multiplex cytokine profiling.

METHOD

- Prospective observational study in a tertiary care cancer center in South India.
- PBMCs from newly diagnosed AML (n = 42) were collected pre-treatment.
- Patients were stratified based on: Clinical response [Complete remission (CR) (n = 16) vs. No complete remission (No CR) (n = 26)] and Serum Vitamin D levels [High (>20 ng/mL, n = 23) vs. Low (<20 ng/mL, n = 19)].
- A 13-color flow cytometry (BD FACSAria III) was used to quantify T cell subsets, including CD4+, CD8+, PD1+, GZB+, T effector memory (TEM), T central memory (TCM), T effector (TEF), naïve T cells, and T regulatory cells (Tregs). Frequencies are presented as percent expression and MFI (Mean Fluorescence Intensity). Data are presented as median (range).
- Cytokines were measured via a 20-plex Bio-Plex assay.
- Data were analyzed using FlowJo, GraphPad Prism, and R. Mann–Whitney U and Spearman correlation tests were applied, with $p < 0.05$ considered significant.



RESULTS

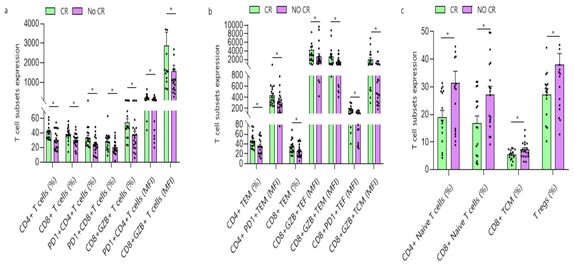
A. Immune Correlates of Clinical Remission (CR vs No CR)

Patients achieving CR showed significantly elevated levels of:

- CD4+ T cells (%): 38.8 vs 28.1; p = 0.018
- CD8+ T cells (%): 36.7 vs 33.0; p = 0.029
- CD8+ GZB+ T cells (%): 48.5 vs 34.8; p = 0.025
- PD1+ CD4+ T cells (MFI): 154 vs 106; p = 0.038

Effector-memory phenotype enhancement in CR:

- CD4+ TEM (%): 43.1 vs 34.6; p = 0.045
- CD8+ TEM (%): 32.8 vs 23.8; p = 0.050
- CD8+ TCM+ GZB+ (MFI): 977 vs 677; p = 0.032
- CD8+ TEM+ GZB+ (MFI): 2504 vs 1040; p = 0.033
- CD8+ TEF+ PD1+ (MFI): 130 vs 88.2; p = 0.050
- CD8+ TEF+ GZB+ (MFI): 3077 vs 1684; p = 0.029



B. Immune Phenotypes Associated with High Vitamin D

Patients with high serum vitamin D demonstrated:

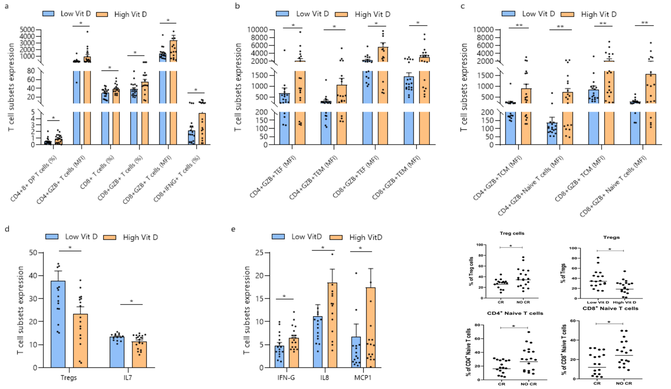
Higher cytotoxic responses:
CD8+ GZB+ (%): 47.1 vs 33.5; p = 0.034
CD4+ GZB+ (MFI): 428 vs 195; p = 0.010
CD8+ GZB+ (MFI): 2336 vs 1137; p = 0.033
CD8+ Naive+ GZB+ (MFI): 1031 vs 254; p = 0.024

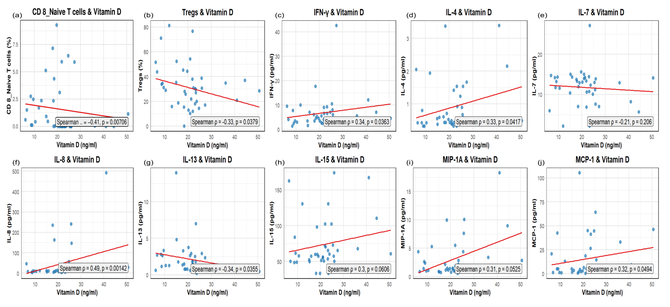
Memory GZB+ enhancement:
CD4+ TCM+ GZB+ (MFI): 475 vs 199.5; p = 0.026
CD4+ TEM+ GZB+ (MFI): 552 vs 250; p = 0.007
CD8+ TCM+ GZB+ (MFI): 1400 vs 626; p = 0.040
CD8+ TEM+ GZB+ (MFI): 2649 vs 1112; p = 0.033
CD8+ TEF+ GZB+ (MFI): 4799 vs 1852; p = 0.024
CD4+ TEF+ GZB+ (MFI): 904 vs 383; p = 0.033

Cytokine elevations:
IFN-γ: 6.28 vs 4.26; p = 0.044
IL-8: 15.64 vs 9.44; p = 0.010
MCP-1: 6.09 vs 2.93; p = 0.019

C. Naive/Treg Skewing in No CR and Low Vitamin D

Non-responders and low-vitamin-D groups showed enrichment of Tregs and naive CD4⁺/CD8⁺ T cells, indicating an immunosuppressive profile.





CONCLUSIONS

AML patients achieving remission exhibit an immune phenotype inclined toward memory-effector differentiation and elevated cytotoxicity. Vitamin D appears to potentiate this effect, suggesting an immunomodulatory role that could inform risk stratification and adjunctive therapy development. These findings support immune–vitamin D interactions as potential biomarkers of clinical response in AML.

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