Characterization of Circulating Large Granulocyte Lymphocytic Proliferation in Patients with Cutaneous T-cell Lymphoma

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Background:

T cell large granular lymphocytes (LGL) are cytotoxic CD8+ lymphocytes, morphologically characterized by moderate amount of cytoplasm with azurophilic and cytotoxic granulation. Its proliferation has been observed in a clinical spectrum from reactive polyclonal expansion to asymptomatic monoclonal expansion to clinical malignant large monoclonal expansion in leukemic phase. T-cell large granular lymphocytic leukemia (LGLL) is a rare hematologic malignancy and manifesting with cytopenia, splenomegaly and autoimmune disorders. Reactive clonal LGL expansions have been described in elderly subjects, viral infections (most commonly, CMV viremia), Felty syndrome (clinical constellation of rheumatoid arthritis, splenomegaly, neutropenia), MDS, post allo-HCT, immunotherapy and autoimmune disease such as Crohn's Disease. However, the association between LGL proliferation and immune dysregulation, causative or epiphenomenon as a consequence of chronic antigen stimulation, is still unclear

Mycosis fungoides/Sezary syndrome (MF/SS) is the most common type of cutaneous T cell lymphoma (CTCL) derived from mature CD4+ T cells and clinically presenting with cutaneous lesions and lymph node and blood involvement in more advanced stage.

Both disorders share common pathogenic pathways such as IL-15/IL-15R. We have recently reported a case of Mycosis Fungoides with CD4+ large granular lymphocytic proliferation mimicking Sezary syndrome (Zhang et al. 2017). The purpose of this study was to expand our understanding of a role of LGL proliferations in patients with CTCL.

Methods:

We searched Moffitt Cancer Center CTCL and flow cytometry databases from 2012 to 2017 for patients with pathologically confirmed diagnosis of CTCL, who also had associated LGL proliferations detected with multicolor flow cytometry. A retrospective chart review was performed on the selected patients.

Clinical and laboratory characteristic were collected and analyzed using descriptive statistical methods. Flow cytometry and T-cell receptor gene rearrangement by PCR of those selected patients were reviewed and confirmed. A subset of the samples from patients with circulating LGL were analyzed for the wavelength of T-cell receptor peak from peripheral blood, tissue sample and bone marrow biopsy to assess for shared clonality between LGL population and CTCL T cell population.

Categorical variables were summarized using counts and percentages.

Continuous variables were summarized using medians with ranges.

Results:

CTCL clinical features

Seventeen patients (N=17) with CTCL demonstrating LGL proliferations were identified. Median age was 55 years [20-82], M:F ratio is 0.88, 71% (12) Caucasian, 18% (3) (African-American) and 11% (2) Hispanic. Among them, 76% (13) had MF, 6% (1) had SS and 23% (3) had other types of CTCL. 93% (15) had early stage (I/IIA) disease.

LGL proliferation features

LGL proliferations were identified after CTCL diagnosis in all patients with a median time of 4 years. Median absolute LGL count was $0.148 \times 10^9/L$. 94% (16/17) patients revealed characteristic LGL immuno-phenotype: CD3+/CD8+/CD57+CD5dim. Among them, 30% (5) showed the secondary CD4+/CD57+ LGL populations. 35% (6) patients showed resolution of LGL population during their follow-up. Median time to resolution was 146 days. 76% (13) had TCR gene rearrangement studies done on bone marrow biopsy, among which 77% (10) had positive results. All 10 patients demonstrated identical TCR gene peaks in bone marrow and peripheral blood. 35% (6) CTCL patients were diagnosed with other malignancies including 18% (3) patients with LGL leukemia. No patient with LGL proliferations developed cytopenia requiring systematic therapy. 35% (6) patients had hepatomegaly, splenomegaly, or lymphadenopathy including 3 pts with LGL leukemia.

Clonality of LGL proliferation and CTCL

8/11 CTCL patients with available TCR gene rearrangement results from skin biopsy revealed clonal T-cell population. Among them, 5 patients had available both TCR gene rearrangement of skin biopsy and of peripheral blood. 2/5 patients showed identical TCR peaks suggesting a possibility of putative mutation in T cell progenitor cell resulting in common pathogenic pathway. In both patients no LGL cells were identified in skin and no CTCL cells were circulating in peripheral blood.

Conclusion:

- Clonal LGL proliferations in CTCL are heterogeneous ranging from benign transient reactive clones to LGLL.
- LGL proliferation including patients with confirmed diagnosis of LGLL likely suggests a benign course since no patients required therapy for LGL proliferations.
- Interestingly, two patients revealed identical TCR gene rearrangement peaks in mycosis fungoides and LGL cells suggesting a possibility of common putative mutation in progenitor T-cell.

Although this is first study to characterize LGL proliferation in CTCL patients, further follow up is needed to compare the clinical course of CTCL patients with and without LGL proliferation to see if a protective role of LGL proliferation can be described. Additionally, translation study is needed to study the putative pathway between CTCL patients with circulating LGL proliferation possibly from same progenitor T cells.

Peripheral blood smear showed many circulating T-cell large granular lymphocytes (Wright stain, $\times 100$). (Zhang et al. 2017).

T cell receptor gene rearrangement detected by PCR and wavelength of TCR was noted next to it. (Zhang et al. 2017).