SciMedCentral

Journal of Hematology & Transfusion

Research Article

Large Granular Lymphocytic Leukemia: Common Therapies and their Outcomes

Manuel G Afable II[1], Michael J Clemente[1], Andres Jerez[1], Li Zhang[2], Holleh Husseinzadeh[1], Hanna Rajala[3], Satu Mustjoki[3], Hideki Makishima[1], Brittney E Dienes[1], John Desamito[4], Alan Lichtin[4], Mikkael A Sekeres[1,4], Brad Pohlman[4], Anjali Advani[4], Ramon V Tiu[1,4] and Jaroslaw P Maciejewski[1,4]*

[1]*Department of Translational Hematology and Oncology Research, Taussig Cancer Institute, USA*

[2]*Department of Quantitative Health Sciences, Cleveland Clinic, Cleveland Clinic, USA*

[3]*Department of Medicine, Hematology Research Unit, Helsinki University Central Hospital, Finland*

[4]*Department of Hematologic Oncology and Blood Disorders, Cleveland Clinic, Cleveland, USA*

Corresponding author

Jaroslaw P Maciejewski, Department of Translational Hematology and Oncology Research, Taussig Cancer Institute, Cleveland Clinic, Cleveland, OH, 9500 Euclid Avenue Cleveland OH USA 44195; Tel: 216-445-5962; Fax: 216-636-2498; Email: maciejj@ccf.org

Submitted: 02 February 2014

Accepted: 18 February 2014

Published: 05 June 2014

ISSN: 2333-6684

Keywords

- T-LGL
- NK-LGL
- STAT3 mutation

Abstract

Large Granular Lymphocytic Leukemia (LGLL), a rare lymphoproliferative disorder of T cell or Natural Killer cell (NK) origin is characterized by persistent immunodominant clones of CD3+ Cytotoxic T Lymphocyte (CTLs) or CD3- NK cells. Dysregulated homeostasis of CTL and NK cells may be due to various pathogenic factors, including the recently identified somatic STAT3 mutations. Occurrence of severe cytopenias necessitates therapy initiation. Due to the rarity of LGL, most of the clinical experience is empiric and based on retrospective analyses. We studied the efficacy of the most frequently used agents in a clinically and molecularly annotated cohort of 152 LGLL patients. Our results demonstrate comparable primary and overall responses to oral cyclosporine A (CsA), Cyclophosphamide (CPM) and Methotrexate (MTX) regimens although the indications spectrum varied due to specific toxicities. Presence of STAT3 mutations did not affect responses or overall survival.

INTRODUCTION

Large Granular Lymphocytic Leukemia (LGLL) is a chronic lymphoproliferative disorder of T cell or NK cell origin characterized by persistent immunodominant clones of CD3+ Cytotoxic T Lymphocyte (CTL) or CD3- Natural Killer (NK) cells. LGLL occurs in elderly patients and comprises 2-5% of chronic lymphoproliferative diseases [1,2]. After antigen clearance, CTLs undergo activation-induced apoptosis, a process that appears dysregulated in LGL [3] due to activation of IL-6 signaling pathways by STAT3 mutations, for example [4]. While T-LGL is often chronic [5], clinical sequelae occur in approximately 50% of patients and necessitate treatment. Neutropenia and hemolytic anemias are the most common presentations, occurring in 61% and 24% of patients, respectively [2] while 25-50% of patients present with splenomegaly [1,6]. Mechanisms purported to be responsible for development of cytopenia include deregulated Fas/Fas ligand-mediated or direct cytotoxic destruction of lineage-committed progenitors, splenomegaly, or cytokine-driven inhibition of maturation [7,8].

As the inherent rarity of this disease precludes the successful conduct of prospective, randomized clinical trials to identify optimal therapies, we conducted a retrospective analysis to compare the therapeutic effectiveness of the most commonly used agents in LGLL.

MATERIALS AND METHODS

Patients

Informed consent was obtained from all study patients for blood and bone marrow aspirate collection and retrospective chart review. The diagnosis of T-LGLL was established using the modified 2008 WHO criteria [9]. Laboratory workup included peripheral blood smear evaluation, flow cytometry with TCR Vb typing (size of dominant VB clone) and TCR gene rearrangement. Although an LGL count >2 x 10^9/L was considered for diagnosis,

patients with lower LGL count were included if other diagnostic criteria were met and the clinical presentation was consistent with LGLL [1]. Clinical data included patient age, gender, splenomegaly (spleen size determined by ultrasound), associated autoimmune conditions, and underlying solid tumor or hematologic malignancies. Cytopenias were classified as follows: neutropenia, absolute neutrophil count (ANC) <1 x 10^9/L; anemia, hemoglobin <10 g/dL; and thrombocytopenia, platelet count <100 x 10^9/L. Hematologic response was defined as Complete Response (CR) when all affected lineages in blood normalized. Partial Response (PR) is defined as improvement in blood counts in the absence of CR. For *STAT3* mutations, amplicon-based deep sequencing and amplification refractory mutation system PCR were performed as previously described [4].

RESULTS AND DISCUSSION

The choice of initial and subsequent therapies in LGLL is empiric as neither factors predicting therapy response nor treatment guidelines have been established. LGL is derived from slow-cycling aberrant memory cells serving as progenitors for the bulk of mature effector CTL (15). Thus, chronic, lower-dose treatments rather than high-dose pulse modalities are used. Nevertheless, some patients referred have previously been treated with high-intensity or pulse. Regimens such as fludarabine, cyclophosphamide+hydroxydaunorobucin+oncovin +prednisone (CHOP), Cyclophosphamide+vincristine+predniso ne (CVP).

Therapy responses

We identified a cohort of 169 patients with presumed LGLL (average disease duration 67 months; 0.26-316); 17 patients (not fulfilling criteria) were excluded. T-LGLL was diagnosed in 88% of patients, chronic NK-LGLL in 10% and aggressive NK-LGLL in 2% (Table 1) of whom 25% had *STAT3* mutations. Rheumatoid arthritis and monoclonal gammopathy of unknown significance / multiple myeloma were the most frequent diseases associated, 15% and 21%, respectively. Indications for treatment were: anemia (49%), neutropenia (37%), mixed (8%) and others (6%). Treatment was required in 85 (56%) patients. As initial treatment, 34/85 (40%) LGLL patients were treated with CsA/ tacrolimus, 22 (26%) with MTX, 18 (21%) with oral CPM and 11 (13%) with other chemotherapeutic agents. Initial response rates (RR) were: CsA 14/28 (50%); CPM 9/16 (56%); MTX 9/20 (45%); Figure 1A) [p=.77]. The median response duration (months) were 24 (3-63), 19 (3-71) and 18 (2-41), respectively (p=.67). Due to lack of initial response and/or toxicity, many patients received multiple therapies. The overall response rates (ORR after 3 combined modalities when the first was CsA, CPM or MTX) were 52%, 50%, and 49%, respectively (Figure 1A) [p=.97]. In our institution, CPM was not commonly used as primary treatment in patients with severe neutropenia nor was MTX used in patients with severe anemia. . We also analyzed RR based on indication for treatment: the initial RR for CsA, CPM and MTX when used for anemia were 6/12 (50%), 8/13 (62%), and 2/6 (33%), respectively (p=.52). For neutropenia, the RR were 8/12 (67%), 0/2, 4/10 (40%), respectively (p=.20) (Figure 1B). In total, 22 patients received low-dose alemtuzumab (10-20 mg SQ/week) as a salvage therapy; ORR in this group was 30%. ORR in patients who underwent splenectomy was 52% with median response duration of 30 months (1.8-115 months).

Table 1: Patient Characteristics.

Parameter	Number
Median age in years (range)	63 (17-87)
Gender (M/F)	87/65
Average duration of disease (mos)	67 (0.26-316)
Diagnosis	
T-LGL	134
NK-LGL, chronic	15
NK-LGL, aggressive	3
Median LGL count, x10^9/L	2410
Median ANC, x10^9/L (range)	1.49 (0-11)
Median PLT count, x10^6/mL (range)	186 (6-627)
Median ALC , x10^9/L (range)	2.84 (0.25-27)
Hematologic manifestation (%) Neutropenia (<1 x10^9/L)	39
Anemia (<10 g/dL)	34
Thrombocytopenia (<100 x10^6/mL)	16
Splenomegaly (%)	28
RBC transfusion dependent (%)	30
Treatment (#)	
CsA/tacrolimus	34
CPM	18
MTX	22
Others*	11

T-LGL: T cell large granular lymphocytic leukemia; NK-LGL: Natural killer cell large granular lymphocytic leukemia; ANC: Absolute Neutrophils Count; PLT: platelet. Alc: Absolute Lymphocyte Count. CsA: Cyclosporine; CPM: Cyclophosphamide; MTX: Methotrexate.
*2 CVP; 2 CHOP/R-CHOP; 1 chlorambucil+prednisone; 1 etoposide+CTX and 5 others.

Retrospective analysis of 374 reported patients across multiple studies (104 with CsA, 148 MTX and 68 CPM) showed a primary RR of 57%, 46% and 59%, respectively [2,8,11-15]. Our results were comparable with primary RR for CsA (50%) by Battiwalla [14]. Osuji reported a higher RR (78%) [5] while in a series of 6 patients only 17% RR was reported [2]. For CPM, our RR (56%) was lower than reported by other authors (69%-75% RR) [2,5,11]. MTX as an initial treatment shows the most variable results across reported studies.

Earlier small studies (<10 patients) reported a primary RR of 60-86% [8,12,13]. Our result was comparable to several previous reports (33% - 44%) [2,15,16].

Laboratory correlates

The choice of first-line agent in LGLL takes into consideration potential toxicities. For example, the presence of renal failure precludes the use of CsA, whereas CPM would be avoided as a chronic therapy (younger patients) due to the risk of secondary malignancies. Helpful guidelines regarding splenectomy include splenomegaly, anemia with adequate reticulocyte count and hemolytic anemia or lack of response to therapy.

Various laboratory parameters may be predictors of clinical

A

Drug			Response rate	
Primary		Overall*	Primary (MT)	Primary (WT)
(%)		(%)	(%)	(%)
Cyclosporine/Tacrolimus	14/28 (50)	31/60 (52)	6/11 (54)	8/17 (47)
Cyclophosphamide	9/16 (56)	15/30 (50)	3/3 (100)	5/12 (42)
Methotrexate	9/20 (45)	18/37 (49)	2/5 (40)	4/10 (40)
Alemtuzumab	N/A	6/20 (30)	N/A	N/A
Others	7/12 (58)	-	N/A	N/A

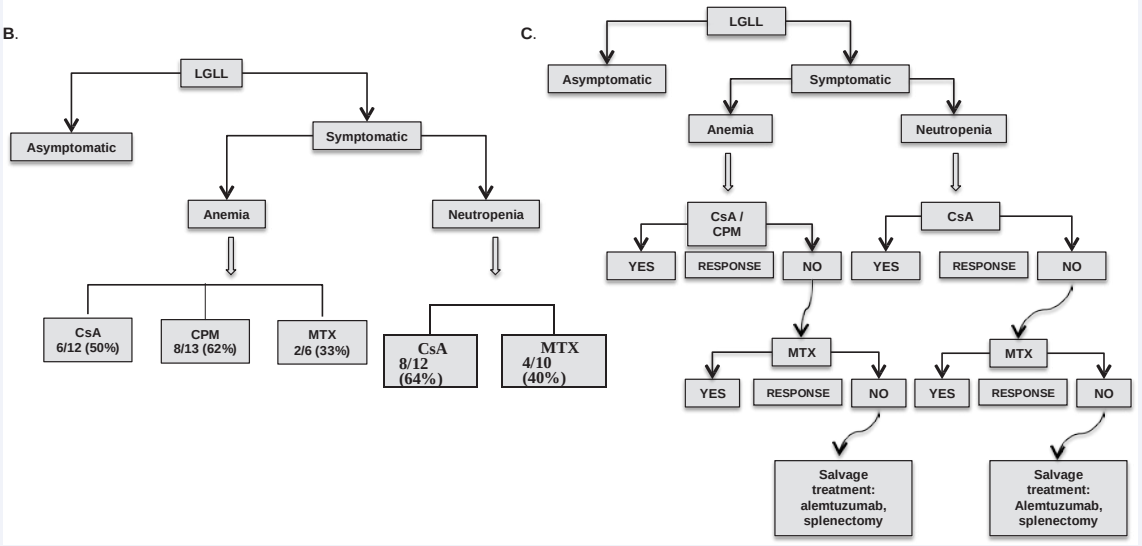

Figure 1 A) Primary response rate and overall response rate. RR: Response Rate; CsA/Tac, cyclosporine/tacrolimus. CPM, cyclophosphamide. MTX, methotrexate. MT, STAT3 mutation. WT, STAT3 wild type. B) Initial response rate based on indication for treatment. CsA: Cyclosporine; CPM: Cyclophosphamide; MTX: Methotrexate. C) Proposed algorithm for treatment of LGL. CsA: Cyclosporine. CPM: Cyclophosphamide. MTX: Methotrexate.
*Response rate after 3 combined modalities. MT , positive for STAT3 mutation. WT, negative for STAT3 mutation.

outcomes. Somatic STAT3 mutations, while more frequent in males (32 vs.14%, p=.03), did not correlate with severity of anemia (p=.57), neutropenia (p=.83), presence of splenomegaly (p=.43), or absolute LGL count (p=.21) and presence of a specific HLA type. No significant difference in RR (57 vs. 48%, p=.65) or OS (p=.39) was found between STAT3 mutant and wild type cases. STAT3 mutant patients had a lower prevalence of MGUS (10% (3/29) vs. 39% (22/75), p=.04), but not of other B-cell dyscrasias (p=.91). Having a STAT3 mutation also did not impact the presence of concomitant malignancies (26 vs. 32%, p=.67), autoimmune conditions (26 vs. 20%, p=.70) or splenomegaly (39 vs. 29%, p=.43). HLA-DQB1 03 was more frequent in responders (40 vs. 24%, p=.09) while HLA-DRB1 03 was less common (11 vs. 25%, p=.06).

CONCLUSION

In summary, our results demonstrate that, aside from MTX (commonly used as initial treatment), comparable response rates can be achieved with CsA and CPM. There is also comparable cross salvage rate among these agents. Although conducted retrospectively, response rates of CPM and CsA have not been systematically presented in a comparative fashion in such large patient group. Our cohort is a well-annotated group of patients seen over a long period with sufficient follow up. Future therapies for LGLL may include an antibody against IL15, previously shown to be upmodulated in T-LGLL [17]. Similarly, IL-6 has also been shown to be overexpressed in LGLL [18] and tocilizumab, an anti-IL6R monoclonal antibody approved for severe RA, is another potentially beneficial therapeutic.

ACKNOWLEDGEMENT

This work was supported by NIH R01 HL082983 (JPM), U54 RR019391 (JPM), K24 HL077522 (JPM), R01 CA113972 (JPM), LLS 624-13 (JPM).

REFERENCES

1. Lamy T, Loughran TP Jr. How I treat LGL leukemia. Blood. 2011; 117: 2764-2774.
2. Bareau B, Rey J, Hamidou M, Donadieu J, Morcet J, Reman O, et al. Analysis of a French cohort of patients with large granular lymphocyte leukemia: a report on 229 cases. Haematologica. 2010; 95: 1534-1541.
3. Zhang R, Shah MV, Yang J, Nyland SB, Liu X, Yun JK, et al. Network model of survival signaling in large granular lymphocyte leukemia. Proc Natl Acad Sci U S A. 2008; 105: 16308-16313.
4. Jerez A, Clemente MJ, Makishima H, Koskela H, Leblanc F, Peng Ng K, et al. STAT3 mutations unify the pathogenesis of chronic lymphoproliferative disorders of NK cells and T-cell large granular lymphocyte leukemia. Blood. 2012; 120: 3048-3057.
5. Osuji N, Matutes E, Tjonnfjord G, Grech H, Del Giudice I, Wotherspoon A, et al. T-cell large granular lymphocyte leukemia: A report on the

treatment of 29 patients and a review of the literature. Cancer. 2006; 107: 570-578.

6. Subbiah V, Viny AD, Rosenblatt S, Pohlman B, Lichtin A, Maciejewski JP. Outcomes of splenectomy in T-cell large granular lymphocyte leukemia with splenomegaly and cytopenia. Exp Hematol. 2008; 36: 1078-1083.

7. Epling-Burnette PK, Loughran TP Jr. Survival signals in leukemic large granular lymphocytes. Semin Hematol. 2003; 40: 213-220.

8. Lamy T, Liu JH, Landowski TH, Dalton WS, Loughran TP Jr. Dysregulation of CD95/CD95 ligand-apoptotic pathway in CD3(+) large granular lymphocyte leukemia. Blood. 1998; 92: 4771-4777.

9. Swerdlow S, Campo E, Harris NL, Jaffe E, Pileri S, Stein H. WHO classification of Tumours of Haematopoietic and Lymphoid Tissues (International Agency for Research on Cancer, Lyon, France). 2008.

10. Melenhorst JJ, Sorbara L, Kirby M, Hensel NF, Barrett AJ. Large granular lymphocyte leukaemia is characterized by a clonal T-cell receptor rearrangement in both memory and effector CD8(+) lymphocyte populations. Br J Haematol. 2001; 112: 189-194.

11. Dhodapkar MV, Li CY, Lust JA, Tefferi A, Phyliky RL. Clinical spectrum of clonal proliferations of T-large granular lymphocytes: a T-cell clonopathy of undetermined significance? Blood. 1994; 84: 1620-1627.

12. Loughran TP Jr, Kidd PG, Starkebaum G. Treatment of large granular lymphocyte leukemia with oral low-dose methotrexate. Blood. 1994; 84: 2164-2170.

13. Hamidou MA, Sadr FB, Lamy T, Raffi F, Grolleau JY, Barrier JH. Low-dose methotrexate for the treatment of patients with large granular lymphocyte leukemia associated with rheumatoid arthritis. Am J Med. 2000; 108: 730-732.

14. Battiwalla M, Melenhorst J, Saunthararajah Y, Nakamura R, Molldrem J, Young NS, et al. HLA-DR4 predicts haematological response to cyclosporine in T-large granular lymphocyte lymphoproliferative disorders. Br J Haematol. 2003; 123: 449-453.

15. Mohan SR, Clemente MJ, Afable M, Cazzolli HN, Bejanyan N, Wlodarski MW, et al. Therapeutic implications of variable expression of CD52 on clonal cytotoxic T cells in CD8+ large granular lymphocyte leukemia. Haematologica. 2009; 94: 1407-1414.

16. Loughran TP, Jr., Yao X, Bennett JM, Litzow MR, Evens AM, Tallman MS. Results of a Prospective Multicenter Phase II Study of Initial Treatment with Methotrexate In LGL Leukemia (ECOG Protocol E5998). ASH Annual Meeting Abstracts. 2010; 116: 702.

17. Chen J, Petrus M, Bamford R, Shih JH, Morris JC, Janik JE, et al. Increased serum soluble IL-15RÎ± levels in T-cell large granular lymphocyte leukemia. Blood. 2012; 119: 137-143.

18. Shvidel L, Duksin C, Tzimanis A, Shtalrid M, Klepfish A, Sigler E, et al. Cytokine release by activated T- cells in large granular lymphocytic leukemia associated with autoimmune disorders. Hematol J. 2002; 3: 32-37.

Cite this article

Afable II MG, Clemente MJ, Jerez A, Zhang L, Husseinzadeh H, et al. (2014) Large Granular Lymphocytic Leukemia: Common Therapies and their Outcomes. J Hematol Transfus 2(2): 1017.