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CASE REPORT

Rituximab efficacy for minimal residual disease eradication in philadelphia-negative acute lymphoblastic leukemia CD20+

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Abstract
Rituximab has never been investigated in MRD persistence/relapse. We describe a clinical picture of a woman with Ph-negative B-ALL CD20$^+$, ineligible to allogeneic transplant, achieving MRD eradication using rituximab monotherapy after frontline chemotherapy and blinatumomab failure. This demonstrates rituximab's potential in MRD$^+$ Ph-negative B-ALL CD20$^+$, warranting further clinical investigation.

Keywords MRD · Rituximab · Lymphoblastic · Leukemia · Eradication

Introduction

Acute lymphoblastic leukemia (ALL) is a rare blood cancer in adults. While pediatric-like intensive chemotherapy has significantly improved survival rates in younger patients, factors like age, biological characteristics, and minimal residual disease (MRD) status influence treatment strategies, including eligibility for allogeneic hematopoietic stem cell transplantation (HSCT), and predicts survival [1–5].

Approximately 30–50% of B-cell ALL (B-ALL) cases express CD20, which is associated with poorer outcomes due to higher relapse rates [6, 7]. However, adding rituximab, an anti-CD20 monoclonal antibody (MoAb), to frontline standard chemotherapy has improved progression-free survival [8]. Despite advances in treatment, many patients with relapsed/refractory (R/R) B-ALL still have poor prognoses. Novel therapies such as inotuzumab ozogamicin (InO), blinatumomab, and CAR-T cells have improved outcomes, even if cure is still not definitely granted [9–11].

This report presents the first documented case of a Philadelphia-negative (Ph) CD20$^+$ B-ALL patient, ineligible for HSCT, who achieved complete MRD eradication with rituximab monotherapy after multiple MRD relapses following chemotherapy and immunotherapy.

Data were collected in accordance with Declaration of Helsinki and Fondazione IRCCS Ca' Granda Ospedale Maggiore Policlinico internal ethic statement guidance. Rituximab off-label administration was approved by the Fondazione IRCSS Ca' Granda Ospedale Maggiore Policlinico ethics committee and patient provided written informed consent.

Case description

In 2019 a 54-year-old woman was diagnosed Ph-negative B-ALL, B-common, CD20$^+$ (bright expression), CD19$^+$, CD22$^+$, according to WHO 2016 and EGIL classification respectively, presenting with hyperleukocytosis (white blood count, 126×10^9). Personal medical history was significant for heart failure with reduced ejection fraction (HFrEF), class NYHA II, secondary to dilatative cardiomyopathy, atrial fibrillation, sever obesity and non-alcoholic fatty liver disease (NAFLD).

She was enrolled in the Italian GIMEMA LAL2317 clinical trial and treated with blinatumomab in association with standard intensive pediatric-like chemotherapy. She experienced severe complications, among which PEG-asparaginase related G3 hepato-toxicity and G4 for hyperbilirubinemia (alanine aminotransferase 430 U/L, total bilirubin 17,7 mg/dl). Complete remission (CR) MRD$^+$

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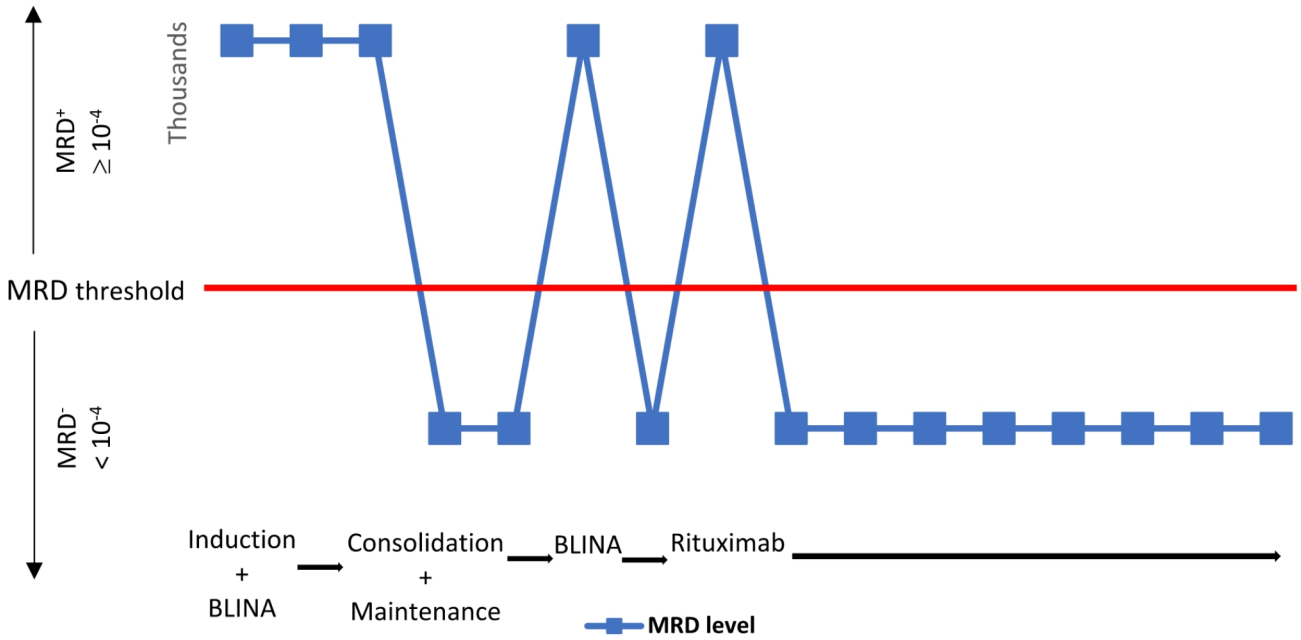

Fig. 1 Minimal residual disease levels during treatment protocols. MRD, minimal residual disease; BLINA, blinatumomab

(10^{-3}), detected by PCR immunoglobulin and/or T-cell receptor gene rearrangements, was reached after three cycles of induction chemotherapy. MRD was eradicated after blinatumomab administration, and she proceeded to the scheduled eight cycles consolidation chemotherapy and one more blinatumomab course as per protocol. HSCT was not feasible because of NAFLD and HFrEF. MRD negativity ($<10^{-4}$, MRD^-) persisted until 23rd maintenance cycle when molecular relapse was detected. Blinatumomab was re-administered in label for total five cycles obtaining a second MRD^- until May 2023 when it turned detectable at low level ($>10^{-4}$). Rituximab was started in an off-label regimen, given $CD20^+$ at diagnosis, with early molecular MRD response after 1 course. She received six courses of anti-CD20 MoAb, once every 21 days.

MRD has been monitored monthly before the start of every subsequent cycle, resulting persistently negative ($<10^{-4}$). Therapy was stopped when she experienced a very severe SARS-COV-2 pneumonia determining admission to intensive care unit. At the last follow-up in February 2025, patient is alive with undetectable MRD after 15 months off therapy (Fig. 1).

Discussion

The patient's clinical history was characterised by PEG-asparaginase hepatotoxicity precluding its further administration, persistent MRD following induction chemotherapy, subsequent relapses, and ineligibility for HSCT, all factors associated to poor prognosis [12–15].

In particular, systematic data confirm that MRD is a key prognostic marker in ALL, with MRD^- strongly correlating with improved progression-free survival (PFS) and overall survival (OS). This strongly suggests the need for a preemptive therapeutic approach to optimize cure rates [16].

In the BLAST study, blinatumomab has demonstrated significant efficacy in prolonging OS and PFS by achieving MRD negativity. Data from a cohort of 116 patients with MRD^+ CR after 3 cycles of intensive chemotherapy were analyzed. Patients received at least one cycle of blinatumomab, after which they either proceeded to HSCT consolidation or continued blinatumomab treatment for up to four cycles, based on the physician's decision. The primary endpoint was MRD eradication after 1 cycle of blinatumomab, attained in 78% of patients. Achieving MRD negativity was associated with longer OS and PFS compared to those MRD^+ (median PFS: 23.6 months vs. 5.7 months; median OS: 38.9 months vs. 12.5 months). Most patients proceeded to HSCT consolidation after blinatumomab, without a clear advantage in terms of relapse free survival., Interestingly, a small group of patients remained in long-term remission without HSCT consolidation [17]. Nevertheless, relapse can still occur, and the median survival for patients who fail blinatumomab has been estimated to be 5.2 months [18].

InO is under investigation in CR MRD^+ patients setting with not definitive results. A phase 2 study reported overall MRD response rates of 69% in 26 patients, with

PFS and OS of 54% and 60% at a median follow-up of 24 months [19]. On the other hand, the interim analysis of the GIMEMA LAL2418 trial reported a MRD eradication rate of 37%, possibly associated with a lower number of InO cycles administered [20].

In addition, CD19/CD22 bispecific CAR-T cells are being investigated in the treatment of CR MRD$^+$ patients. A phase 1 study reported an overall MRD response rate of 100% at day 28, with 24-month PFS and OS rates of 77% and 86%, respectively. Five out of fifteen patients (33%) experienced hematological or MRD relapse [21].

The association of rituximab with induction chemotherapy in Ph-negative CD20$^+$ B-ALL has shown remarkable results in adult settings, becoming the standard of care in many countries. In the USA, its combination with modified Hyper-CVAD demonstrated an OS of 75% versus 47% compared with controls at three years [22]. Similarly, a French group randomized 209 adult patients with de novo CD20$^+$ B-ALL to receive intensive chemotherapy with or without rituximab. The primary endpoint was PFS, with estimated two-year PFS rates of 65% in the experimental arm and 52% in the control arm [8]. A pilot randomized study on a small number of pediatric patients suggested a contribution of rituximab to an early deeper blast clearance during induction chemotherapy [23]. However, rituximab's role in eradicating MRD persistence in adult Ph-negative CD20$^+$ B-ALL, remains largely unexplored, even if its potential efficacy has been suggested. A case report described MRD eradication in a Ph-positive CD20$^+$ B-ALL patient that allowed successful bridging to autologous transplant [24]. A small case series has been reported of 3 pediatric R/R Ph-negative CD20$^+$ B-ALL treated with rituximab combined with salvage chemotherapy, allowing the achievement of a second CR. MRD reduction or eradication was attained in all patients, with a bridge to HSCT in two patients [25].

To our knowledge, this is the first reported case demonstrating the efficacy of rituximab as a single agent in eradicating MRD in Ph-negative CD20$^+$ B-ALL.

Improving survival after blinatumomab failure remains an unmet clinical need, particularly in those patients ineligible for HSCT, with no other effective treatment options currently available. InO and CAR-T cell therapies are under investigation for their potential efficacy in the MRD$^+$ setting, and results are eagerly awaited.

Our findings provide a proof of concept for the effectiveness of rituximab in MRD eradication in Ph-negative CD20$^+$ B-ALL, emphasizing the need for validation of its therapeutic efficacy in larger studies and prospective clinical trials.

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Data availability No datasets were generated or analysed during the current study.

Declarations

Competing interests The authors declare no competing interests.

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