3. PROTOCOL SYNOPSYS

Title	Reduced intensity conditioning with high-dose rituximab followed by allogeneic transplantation of hematopoietic cells for the treatment of relapsed/refractory B-cell non Hodgkin's lymphomas
Study type	Prospective, phase II study
Endpoints	**Primary:** • Progression-free survival at one year **Secondary:** • Overall survival • Engraftment • Incidence of acute graft-versus-host disease (aGVHD) • Incidence of chronic graft-versus-host disease (cGVHD) • Nonrelapse mortality at one year • Percentage of molecular remissions at one year for patient having a molecular marker.
Sample size	190 patients
Study design	This study will be performed as a prospective multicenter, phase II trial.
Study duration	Enrollment, planned to be completed by 3 years, will continue up to 2014 plus 1 year minimum follow-up for the last patient enrolled
Selection criteria	**Inclusion criteria** **1.** Age ≥ 18 ≤ 65 years **2.** Histologies as follow: 2a.Chronic lymphocytic leukemia (CLL) or small lymphocytic lymphoma (SLL) relapsing after at least 2 lines of conventional chemotherapy or relapsing after a first line (within one year) including purine analogue-based regimen or relapsing after autologous stem cell transplantation (as first or second line) 2b. Primary refractory CLL or SLL 2c.Follicular lymphomas (FCL) and nodal marginal zone (MZL) lymphoma relapsing after 2 lines or relapsing after autologous stem cell transplantation 2d.Primary refractory FCL and MZL 2e.Mantle cell lymphomas (MCL) relapsing after conventional chemotherapy or autologous stem cell transplantation 2f.Diffuse large B-cell lymphomas (DLBCL) or transformed FCL relapsing after two lines of conventional chemotherapy or autologous stem cell transplantation 2g.CLL, FCL, MCL, MZL and DLBCL considered eligible for high-dose chemotherapy, with a positive bone marrow biopsy or collecting PCR positive harvests before the autografting phase **3.** PS (Karnofsky) ≥ 70% **4.** HLA-identical (A, B, C, DR, DQ loci) or one antigen mismatched (class I) sibling donors. Donor selection is based on molecular high-resolution typing (4 digits) of the HLA gene loci class I (HLA-A, B, and C) and class II (DRB1, DQB1). In case, no class I and class II completely identical donor (10 out of 10 gene loci) can be identified, the degree of histocompatibility between patient and donor must fulfill with the minimal degree of matching established by the Italian Bone Marrow Donor Registry: HLA-A and HLA-B antigen histocompatibility and HLA-DRB1 allelic histocompatibility. **5.** Written informed consent

	Exclusion criteria **1.** Central nervous system localization **2.** Positive serologic markers for human immunodeficiency virus (HIV) **3.** Active hepatitis B virus (HBV) or hepatitis C virus (HCV) infection **4.** Serum bilirubin levels > 2 the upper normal limit **5.** Ejection fraction < 45% (or myocardial stroke in the last year) **6.** Clearance of creatinine < 50 ml/min **7.** DLCO < 50% **8.** Pregnancy or lactation **9.** Patient not agreeing to take adequate contraceptive measures during the study (up to one year after allogeneic transplantation) **10.** Psychiatric disease **11.** Any active, uncontrolled infection **12.** Type I hypersensivity or anaphylactic reactions to rituximab
Treatment plan	**Conditioning regimen for identical sibling or unrelated donor** Rituximab 500 mg/ms (day –6); Thiotepa 6 mg/Kg every 12 hours for 2 doses (day –5); cyclophosphamide 30 mg/kg (days –4 and –3); fludarabine 30 mg/ms (days –4 and –3); allogeneic stem cell transplantation with 4 - 8 X 10^6 $CD34^+$ cells/kg from related donors (day 0). **GVHD prophylaxis:** Cyclosporin A (CSA), adjusted to 200-300 ng/ml blood levels, and short course methotrexate (10 mg/ms day +1, 8 mg/ms day + 3 and +6). CSA is administered at full dose through day +100 and, if GVHD does not occur, the dose is tapered by 10% every week thereafter. Patients with related sibling with a class I antigen mismatch or with unrelated donor will receive rabbit anti-thymocyte globulin (Thymoglobuline 0.5 mg/kg daily on day -4, Thymoglobuline 3 mg/kg on day –3 and Thymoglobuline 3.5 mg/kg– 2)
Study Procedures at baseline	• Medical history and physical examination • Complete blood counts with differential and platelets • Clinical laboratory evaluations • Serum pregnancy test • HIV-Ab, HBsAg, HCV-Ab, CMV-Ab, Toxo-Ab, EBV-Ab • EKG and ultrasound evaluation of left ventricular ejection fraction • Chest x-ray • CT scan total body • PET scan • Bone marrow biopsy • Examination of Waldeyer's ring • Evaluation of DLCO • Neurologic evaluation and analysis of cerebro-spinal fluid when indicated by clinical symptoms • Peripheral blood sample (at least 4 mL) for immunophenotype analysis • Evaluation of Comorbidity score
Statistical considerations	The study sample size was calculated to estimate the 1-year PFS and corresponding 90% one-sided confidence interval. In a retrospective series of 115 patients with relapsed/refractory B-cell non Hodgkin's

	lymphomas, 1-year PFS was 70%, with 20% relapses. Assuming no treatment effect on NRM and a 35% relative improvement of relapse rate (from 20% to 13%), at a significance level of 10% (one-side test) a sample size of 190 assessable patients ensures a 80% probability of detecting a 70% to 77% increase in 1-year PFS.