

## How important is to eradicate minimal residual disease in indolent non-Hodgkin's lymphoma?

A. RAMBALDI

Divisione di Ematologia,  
Ospedali Riuniti Bergamo, Italy

In Follicular non Hodgkin's Lymphoma (FL-NHL) the molecular diagnosis and monitoring of the chimeric BCL2/IgH gene generated by the t(14;18)<sup>1</sup> is an important tool in the diagnostic work-up at diagnosis and during the clinical follow-up.<sup>2</sup> Although the clinical significance of circulating BCL2/IgH<sup>+</sup> cells is still controversial,<sup>3,4</sup> the persistence of these cells after conventional or high dose chemotherapy programs correlates with a shorter clinical remission.<sup>5-6,7,8</sup> Quantitative PCR assays have been developed<sup>9-11</sup> which provide a reliable tool for the accurate evaluation of BCL2/IgH<sup>+</sup> cells in the BM or peripheral blood (PB)<sup>12</sup> and allow a better molecular monitoring of minimal residual disease after different therapeutic protocols. We have recently shown that the quantitative evaluation of the BCL2/IgH chimeric gene performed at diagnosis in the BM and PB can provide useful information as surrogate marker of the clinical outcome of FL-NHL patients.<sup>13</sup> The clearance of neoplastic cells from BM and PB obtained either after conventional chemotherapy or after passive immunotherapy using the anti-CD20 chimeric monoclonal antibody Rituximab, can be accurately quantified. In our experience, an anthracycline containing regimen like CHOP and Rituximab are both able to remove approximately 2 logs of tumor infiltration thus explaining why patients with a limited lymphoma infiltration (1 positive cell in 10<sup>4</sup> normal cell or less) can achieve a molecular remission after CHOP chemotherapy alone. On the other hand, patients showing at diagnosis intermediate or high levels of BM and PB infiltration, benefit from the addition of Rituximab since they achieve a molecular CR in more than 70% of patients. We also confirm the notion that achieving the combined end point of clinical and molecular remission is one of the major goals in the therapy of FL-NHL. Indeed, no matter whether after CHOP alone or after sequential CHOP and Rituximab, patients in complete clinical and molecular remission show a significantly

longer freedom from disease recurrence. In keeping with our results, several investigators have recently provided evidence on the value of RQ-PCR analysis in patients undergoing autologous hematopoietic stem cell transplantation. Ladetto and co-workers showed that the evaluation of tumor burden by RQ-PCR in stem cell harvest can predict the effectiveness of *ex vivo* purging after high dose chemotherapy.<sup>14</sup> Along the same line, it has been shown that the ability of Rituximab to eradicate contaminating tumor cells in the graft can contribute to improve the clinical outcome of autologous transplantation.<sup>15,16</sup> Moreover, the tumor load of BCL2/IgH<sup>+</sup> cells detected in BM or PB samples post-autologous or allogeneic transplantation in FL-NHL was also found to positively correlate with the duration of clinical remission.<sup>17-19</sup> In addition, our results further support the notion that Rituximab should be promptly offered to all patients who fail to achieve a complete clinical and molecular response at the end of front line therapy. Moreover, the quantitative PCR analysis performed at diagnosis may help in defining patients eligible for studies using high dose chemotherapy as initial treatment.<sup>20</sup> In conclusion, our results support the clinical value of a quantitative evaluation of BCL2/IgH<sup>+</sup> cells at diagnosis, which may help to define the probability of response to conventional chemotherapy with or without the addition of Rituximab. Our data also confirm that molecular monitoring of minimal residual disease allows an early identification of patients with a remarkably higher risk of disease recurrence.

### References

1. Gribben JG, Freedman AS, Neuberg D, et al. Immunologic purging of marrow assessed by PCR before autologous bone marrow transplantation for B-cell lymphoma. *N Engl J Med* 1991;325:1525-33.
2. Lopez-Guillermo A, Cabanillas F, McDonnell TI, et al. Correlation of bcl-2 rearrangement with clinical characteristics and outcome in indolent follicular lymphoma. *Blood* 1999;93:3081-7.
3. Price CG, Meerabux J, Murtagh S, et al. The significance of circulating cells carrying t(14;18) in long

- remission from follicular lymphoma. *J Clin Oncol* 1991;9:1527-32.
4. Lambrechts AC, Hupkes PE, Dorssers LC, van't Veer MB. Clinical significance of t(14; 18)-positive cells in the circulation of patients with stage III or IV follicular non-Hodgkin's lymphoma during first remission. *J Clin Oncol* 1994;12:1541-6.
  5. Lopez-Guillermo A, Cabanillas F, McLaughlin P, et al. The clinical significance of molecular response in indolent follicular lymphomas. *Blood* 1998;91:2955-60.
  6. Freedman AS, Neuberg D, Mauch P, et al. Long-term follow-up of autologous bone marrow transplantation in patients with relapsed follicular lymphoma. *Blood* 1999;94:3325-33.
  7. Tarella C, Cuttica A, Vitolo U, et al. High-dose sequential chemotherapy and peripheral blood progenitor cell autografting in patients with refractory and/or recurrent Hodgkin lymphoma: a multicenter study of the intergruppo Italiano Linfomi showing prolonged disease free survival in patients treated at first recurrence. *Cancer* 2003;97:2748-59.
  8. Rambaldi A, Lazzari M, Manzoni C, et al. Monitoring of minimal residual disease after CHOP and rituximab in previously untreated patients with follicular lymphoma. *Blood*. 2002;99:856-862.
  9. Dolken L, Schuler F, Dolken G. Quantitative detection of t(14;18)-positive cells by real-time quantitative PCR using fluorogenic probes. *Biotechniques* 1998;25:1058-64.
  10. Luthra R, McBride JA, Cabanillas F, Sarris A. Novel 5' exonuclease-based real-time PCR assay for the detection of t(14;18)(q32;q21) in patients with follicular lymphoma. *Am J Pathol* 1998;153:63-8.
  11. Sanchez-Vega B, Vega F, Hai S, Medeiros LJ, Luthra R. Real-Time t(14;18)(q32;q21) PCR assay combined with high-resolution capillary electrophoresis: a novel and rapid approach that allows accurate quantitation and size determination of bcl-2/JH fusion sequences. *Mod Pathol* 2002;15:448-53.
  12. Summers KE, Davies AJ, Matthews J, et al. The relative role of peripheral blood and bone marrow for monitoring molecular evidence of disease in follicular lymphoma by quantitative real-time polymerase chain reaction. *Br J Haematol* 2002;118:563-6.
  13. Rambaldi A, Carlotti E, Oldani E, et al. Quantitative PCR of bone marrow BCL2/IgH+ cells at diagnosis predicts treatment response and long-term outcome in follicular non-Hodgkin lymphoma. *Blood*. 2005;105:3428-33.
  14. Ladetto M, Mantoan B, Ricca I, et al. Recurrence of Bcl-2/IgH polymerase chain reaction positivity following a prolonged molecular remission can be unrelated to the original follicular lymphoma clone. *Exp Hematol* 2003;31:784-8.
  15. Horwitz SM, Negrin RS, Blume KG, et al. Rituximab as adjuvant to high-dose therapy and autologous hematopoietic cell transplantation for aggressive non-Hodgkin lymphoma. *Blood* 2004;103:777-83.
  16. Mahe B, Milpied N, Mellerin MP, et al. PCR detection of residual Bcl-2/IgH-positive cells after high-dose therapy with autologous stem cell transplantation is a prognostic factor for event-free survival in patients with low-grade follicular non-Hodgkin's lymphoma. *Bone Marrow Transplant* 2003;31:467-73.
  17. Hirt C, Dolken G. Quantitative detection of t(14;18)-positive cells in patients with follicular lymphoma before and after autologous bone marrow transplantation. *Bone Marrow Transplant* 2000;25:419-426.
  18. Chang CC, Bredeson C, Juckett M, Logan B, Keever-Taylor CA. Tumor load in patients with follicular lymphoma post stem cell transplantation may correlate with clinical course. *Bone Marrow Transplant* 2003;32:287-91.
  19. Mandigers CM, Verdonck LF, Meijerink JP, Dekker AW, Schattenberg AV, Raemaekers JM. Graft-versus-lymphoma effect of donor lymphocyte infusion in indolent lymphomas relapsed after allogeneic stem cell transplantation. *Bone Marrow Transplant* 2003;32:1159-63.
  20. Ladetto M, Corradini P, Vallet S, et al. High rate of clinical and molecular remissions in follicular lymphoma patients receiving high-dose sequential chemotherapy and autografting at diagnosis: a multicenter, prospective study by the Gruppo Italiano Trapianto Midollo Osseo (GITMO). *Blood* 2002;100:1559-65.