

accumulation of mutated alleles in MF transformation of PV. Finally, in PET-MF the median allele burden was 57%, a value significantly greater than the 26% of a control group of ET patients from our Institution.<sup>13</sup> This result suggests that accumulation of mutant V617F alleles is also a mechanism of evolution toward MF in JAK2V617F mutated ET patients.

Apart from a significantly lower platelet count ( $p=0.04$ ) in JAK2V617F mutated patients, no other meaningful correlation with JAK2V617F mutational status could be ascertained (Table 1). The V617F mutated or unmutated patients were also comparable in terms of spleen size, incidence of thrombosis, incidence of major hemorrhages (2 patients in each group, all from previous ET), and the rate of AML transformation which occurred in 2/16 JAK2 unmutated and 6/49 JAK2V617F mutated patients (12.5% and 12.2% respectively) (*data not shown*). The correlation between V617F allele burden and clinical presentation and prognosis was also studied by dividing patients into quartiles of V617F allele distribution. We grouped together the first two quartiles because of the low number of patients included (Table 1), and we found that V617F allele burden was positively correlated with age, white blood cell count and circulating CD34<sup>+</sup> cell count (all with a  $p$  value  $<0.05$ ). However, there was no statistically significant correlation with clinical characteristics, i.e. spleen size, thrombosis, hemorrhages, nor with the rate of patients who evolved to AML (3 of whom were in the two first quartiles, one and 2 in the third and fourth quartile respectively).

To the best of our knowledge, this is the first study specifically addressing the clinical relevance of JAK2V617F mutation in patients who evolved to MF from a previous PV or ET. The data presented herein prompted us to conclude that neither the JAK2V617F mutational status nor the V617F allele burden seem to have relevance for disease phenotype or prognosis in this setting of patients. However, the relatively short follow-up might have prevented the discovery of correlations of JAK2V617F mutational status with clinical events occurring late in the history of these disorders, in particular for the evolution to AML. This will require larger patient series with a much longer observation.

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**Key words:** myelofibrosis, JAK2V617F mutation, polycythemia vera, essential thrombocythemia

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**Funding:** the study was supported by Ministero della Università e Ricerca (COFIN 2006067001\_003), Istituto Toscano Tumori, and institutional funds from Università di Firenze (ex-60%).

**Citation:** Guglielmelli P, Barosi G, Pieri L, Antonioli E, Bosi A, Vannucchi AM. JAK2V617F mutational status and allele burden

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doi: 10.3324/haematol.13721

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## High-dose therapy and autologous stem cell transplantation versus conventional therapy for patients with advanced Hodgkin's lymphoma responding to front-line therapy: long-term results

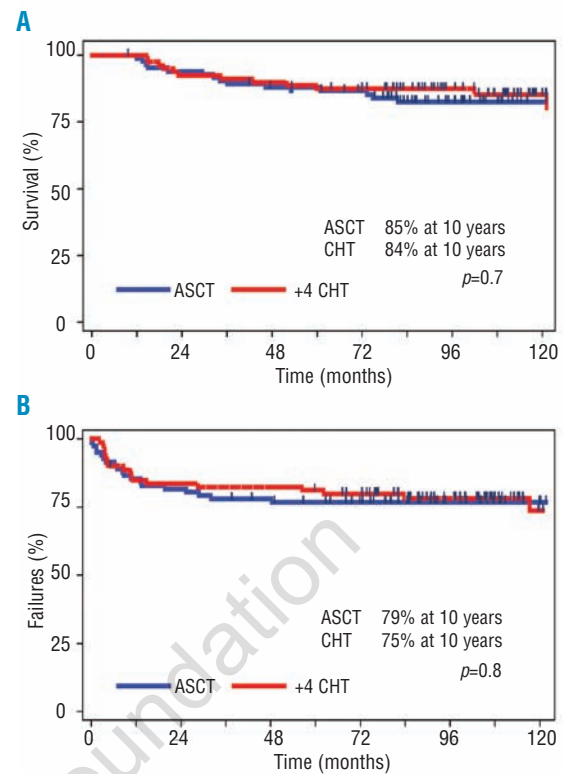
The inclusion of high-dose therapy/autologous stem cell transplantation (HDT/ASCT) in the initial treatment plan for patients with unfavorable Hodgkin's lymphoma

(HL) has been a matter of debate for the last two decades. In 1991, Carella *et al.*<sup>1</sup> published a pilot study of HDT and ASCT in patients with unfavorable HL who had achieved CR with conventional-dose therapy. In an attempt to improve the cure rate of advanced, unfavorable HL and to reduce the risk of relapse after initial response, in April 1993 we started a large co-operative study comparing HDT/ASCT versus conventional chemotherapy (CHT) in patients in CR or PR after four courses of ABVD or other doxorubicin-containing regimens.

Between April 1993 and December 2000, 208 patients were registered on the trial; among these, 163 fully complied with the protocol requests and were randomly assigned to receive HDT plus ASCT (HDT-ASCT Arm, 83 patients) versus four additional courses of the same standard chemotherapy used in the induction phase (CHT Arm, 80 patients). Unfavorable HL was defined using a Strauss-derived system.<sup>2</sup> The details relative to patients' characteristics, entry criteria, treatment protocol and statistical analysis have been previously described.<sup>3</sup> At the end of the treatment program, 92% of patients in HDT-ASCT arm and 89% in CHT arm achieved a CR ( $p=0.6$ ). The 5-year FFS rates were 75% (95% confidence interval [CI], range 65-85) in the HDT-ASCT arm and 82% (95% CI, range 73-90) in the CHT arm ( $p=0.4$ ). The 5-year OS rates were 88% (95% CI, range 80-96) in the HDT-ASCT arm and 88% (95% CI, range 79-96) in the CHT Arm ( $p=0.99$ ). The 5-year RFS rates were 88% in the HDT-ASCT arm (95% CI, range 80-96) and 94% in the CHT arm (95% CI, range 88-100), and there was no statistical difference ( $p=0.3$ ). We now present the updated results of this study after an extended follow-up period of four years (median follow-up of alive patients is currently 107 months, range 9-172). In Table 1 we present a flow diagram showing what became of all the patients assigned to each arm and, finally, 10-year OS, RFS and FFS.

The 10-year OS were 85% (95% CI, range 78-90) and 84% (95% CI, range 77-89) for patients who underwent HDT-ASCT or CHT respectively. Also after an extended follow-up, no significant difference emerged between the two arms ( $p=0.7$ ) (Figure 1A). Since the Hasenclever prognostic score has emerged to be more robust than the Strauss prognostic system, we also performed the analysis of our data according to the former.<sup>4</sup> We compared outcome in the two arms but we found no statistical difference [10 year OS: IPS 0-2 (49 pts.): 98% (ASCT), 84% (CHT),  $p=0.5$ ; IPS 3-6 (84 pts.): 85% (ASCT), 88% (CHT),  $p=0.8$ ; 10 year FFS: IPS 0-2 (49 pts.): 84% (ASCT), 75% (CHT),  $p=0.4$ ; IPS 3-6 (84 pts.): 79% (ASCT), 83% (CHT),  $p=0.8$ ]. The 10-year FFS was 79% (95% CI, range 72-85) for patients in the HDT-ASCT arm and 75% (95% CI, range 67-82) for patients in the CHT arm ( $p=0.8$ ) (Figure 1B). No difference in late toxicity was observed between the two groups: overall late toxicity consisted of 2 bone necrosis in the HDT/ASCT arm and 2 second malignancies, (cancer of the testis with gastric metastases and one a MDS/AML). In addition to these failures, 2 patients have died from a second malignancy (both lung cancers), one in each arm.

Therefore, after a median follow-up of 107 months,



**Figure 1.** (A) Overall survival and (B) failure-free survival of all 163 randomized patients. ASCT: autologous stem-cell transplantation; CHT: chemotherapy.

our data definitely support the view that for patients responding to initial conventional CHT the consolidation with HDT/ASCT is not superior to consolidation with conventional-dose therapy and, most importantly, confirm that HDT/ASCT as consolidation therapy should no longer be offered to these patients.

Another relevant issue of our study was the evaluation of safety of an early use of HDT followed by ASCT. According to published data, 2% of deaths caused by HDT/ASCT toxicity can be expected.<sup>5</sup> In the present study, only one toxic death occurred thus confirming the increasing safety of this procedure when performed in the initial course of the disease. Of course, these conclusions refer to a population diagnosed with HL in the pre-FDG-PET era, when the treatment decisions were based on adverse risk factors present at time of diagnosis, before treatment was started. Currently, we have more chance of reducing treatment related toxicities and of avoiding undue treatment in selected groups of patients who can be cured with standard-dose regimens. Using FDG-PET scan in the evaluation of treatment response in HL, the negative predictive value is high (81-100%) showing the ability of FDG-PET to identify patients with excellent prognosis.<sup>6,8</sup> To these patients we can reasonably apply the conclusions of our study, that is that they do not benefit from an early intensification with HDT and ASCT. The ongoing worldwide trials will most probably provide a definite answer to the role of early PET in the selection of

**Table 1.** Flow diagram showing the updated results of randomized patients.

	HDT/ASCT arm (83)	CHT arm (80)	
Complete remission	76 (92%)	71 (89%)	
Deaths			
Overall	14	12	
During extended follow-up	6	3	
Relapses			
Overall	8	7	
During extended follow-up	–	3	
Failures*			
Overall	20	18	
During extended follow-up	1	4	
Median follow-up, months (alive pts.)	108 (42-172)	106 (9-158)	
10-year OS	85% (95% C.I. 78-90)	84% (95% C.I. 77-89)	<i>p</i> =0.7
10-year RFS	88% (95% C.I. 81-95)	89% (95% C.I. 83-93)	<i>p</i> =0.8
10-year FFS	79% (95% C.I. 72-85)	75% (95% C.I. 67-82)	<i>p</i> =0.8

\*CR not achieved, relapse after CR, death in CR, second tumor.

patients at high risk of relapse candidates for intensification with HDT and ASCT.

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Funding: supported by a grant from "Associazione Angela Serra per la Ricerca sul Cancro," Modena, Italy.

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Citation: Carella AM, Bellei M, Brice P, Gisselbrecht C, Visani G, Colombat P, Fabbiano F, Donelli A, Luminari S, Feugier P, Browett P, Hagberg H, and Federico M. High-dose therapy and autologous stem cell transplantation versus conventional therapy for patients with advanced Hodgkin's lymphoma responding to front-line therapy: long-term results. *Haematologica* 2009; 94:146-148. doi: 10.3324/haematol.13484

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## Clinical relevance of MDM2 SNP 309 and TP53 Arg72Pro in follicular lymphoma

Tumor protein 53 (TP53) is critical to cell cycle control and is the most common mutational target in germinal center lymphomas. However, these mutations occur infrequently at diagnosis (<10%) in follicular lymphoma (FL), and are more commonly associated with disease progression or transformation to more aggressive histology.<sup>1-</sup>

Inactivation of TP53 may also occur by upregulation of the human homolog of the Murine Double Minute 2 protein (MDM2), which targets TP53 for degradation by the proteasome and is frequently amplified in FL and other malignancies; indeed *Mdm2* haplo-insufficiency in mice leads to enhanced TP53 function with delayed onset of lymphoma.<sup>5</sup> While many studies have examined the genomic events targeting these loci, less is known as to the functional role of polymorphic variants at the *MDM2* or *TP53* loci. The single nucleotide polymorphisms (SNPs), *MDM2* SNP 309 and *TP53* Arg72Pro, have at least in some studies an additive effect on cancer susceptibility with *MDM2* SNP 309 also predicting advanced disease at diagnosis.<sup>6</sup> Consequently, we investigated the impact of *MDM2* SNP 309 and *TP53* Arg72Pro on the clinical outcome of FL. *MDM2* SNP 309 characterizes a T>G substitution at nucleotide 309 of intron one and leads to higher *MDM2* mRNA and protein expression with lower apoptotic response. Significantly, homozygosity for the G allele correlates with earlier onset of *de novo* diffuse large B-cell lymphoma (DLBCL) in females.<sup>7</sup> Its role in CLL is less clear as two recent studies provide contrasting results.<sup>8,9</sup> *TP53* Arg72Pro is a non-synonymous SNP involving G>C substitution at nucleotide 466 of exon 4 in