MRD response is potentially influenced by *MRP4* polymorphisms modulating dose intensity of these drugs.

Using this approach, we did not find any important MRD-predisposing *MRP4* genotype. In addition, comparisons of Kaplan-Meier estimates did not detect significant differences in EFS for patients with different *MRP4* genotypes. At most we observed an oppositional trend compared with the study of Ansari toward a poorer EFS for carriers of TC-1393 and AC/CC934 (group 1),[1] whereas patients with TT-1393 and AC934 (group 3)[1] tended toward a better EFS (Figure 1).

The discrepancy cannot be ascribed to differences in genotype distribution between the 2 ALL series (frequencies in this study vs Ansari et al: T-1393C: TT 88.7% vs 88.3%, TC 11.1% vs 11.7%, CC 0.2% vs 0.0%; C934A CC 87.7% vs 85.8%, CA 11.1% vs 13.1%, AA 1.2% vs 1.1%).[1] Ansari et al describe higher frequencies of treatment induced toxicity and higher MTX plasma levels suggesting a higher MTX dose intensity for patients with group 3 *MRP4* genotype. They conclude that this might lead to more frequent drug withdrawal or dose reduction, potentially causing higher frequency of relapse within this group.[1] MTX dose intensity during early induction consolidation is lower in GMALL protocols (MTX dose 1.5 g/m$^2$ within GMALL protocols[2] vs 4 g/m$^2$ within DFCI protocols[7,8]). Therefore, the proposed modulation of MTX dose intensity by *MRP4* genotype may cause other effects than observed within the DFCI protocols. Age difference between analyzed patient series may also influence the prognostic value of specific polymorphisms.

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**This study has been approved by the institutional review board of the University of Frankfurt and was supported by the Wilhelm Sander-Stiftung (grant no. 2001.074.2).**

**Contribution:** M.B. designed the study, contributed to data analysis, and wrote the manuscript; H.T. performed and interpreted molecular analyses; D.H. and N.G. are the chairmen/coordinators of the GMALL trials responsible for the overall clinical conduct of the studies, provided the information on presenting clinical, immunophenotypic, and genetic features and follow-up-data, and contributed to the preparation of the paper; M.K. was responsible for the overall conduct of the MRD study and participated in manuscript preparation; and T.R. analyzed data and contributed to molecular analyses and manuscript preparation.

**Conflict-of-interest disclosure:** The authors declare no competing financial interests.

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## References

1. Ansari M, Sauty G, Labuda M, et al. Polymorphisms in multidrug resistance-associated protein gene 4 is associated with outcome in childhood acute lymphoblastic leukemia. *Blood.* 2009;114:1383-1386.
2. Brüggemann M, Raff T, Flohr T, et al. Clinical significance of minimal residual disease quantification in adult patients with standard-risk acute lymphoblastic leukemia. *Blood.* 2006;107:1116-23.
3. Raff T, Gökbuget N, Lüschen S, et al. Molecular relapse in adult standard-risk ALL patients detected by prospective MRD monitoring during and after maintenance treatment: data from the GMALL 06/99 and 07/03 trials. *Blood.* 2007; 109:910-915.
4. Schwender H, Ickstadt K. Identification of SNP interactions using logic regression. *Biostatistics.* 2008;9:187-198.
5. Russel FG, Koenderink JB, Masereeuw R. Multidrug resistance protein 4 (MRP4/ABCC4): a versatile efflux transporter for drugs and signalling molecules. *Trends Pharmacol Sci.* 2008;29:200-207.
6. van der Velden VH, Cazzaniga G, Schrauder A, et al. Analysis of minimal residual disease by Ig/TCR gene rearrangements: guidelines for interpretation of real-time quantitative PCR data. *Leukemia.* 2007;21:604-611.
7. Dulucq S, St-Onge G, Gagné V et al. DNA variants in the dihydrofolate reductase gene and outcome in childhood ALL. *Blood.* 2008;111:3692-700.
8. Moghrabi A, Levy DE, Asselin B, et al. Results of the Dana-Farber Cancer Institute ALL Consortium Protocol 95-01 for children with acute lymphoblastic leukemia. *Blood.* 2007;109:896-904.

# Response

## *MRP4* gene polymorphisms and treatment response in adult ALL

We thank Brüggemann and colleagues for their interest in our paper[1] that recently reported an association between *MRP4* gene polymorphisms (regulatory T-1393C and A934C leading to Lys304Asn substitution) and outcome in childhood acute lymphoblastic leukemia (ALL) patients treated on Dana-Farber Cancer Institute (DFCI) protocols. Given the preliminary character of this study, replication in an additional cohort of sufficient size was suggested. Brüggemann et al analyzed same polymorphisms in adult ALL patients who underwent treatment with German Multicenter ALL (GMALL) protocols. The cohort was enough powered to detect genotype-associated differences, but nevertheless failed to do so, further confirming little concordance that exist between pharmacogenetics findings in childhood and adult ALL.[2] The authors suggest that the difference in methotrexate (MTX) dose between protocols account for observed discrepancy. While we agree with such a possibility, we would like to emphasize several other reasons possibly contributing to this finding. ALL in childhood and adulthood are distinct diseases as based on the observed difference in etiology, incidence rate, disease characteristics and survival.[3] Disease-free survival in childhood ALL is as high as 80%, whereas it barely reaches 40% in adults. ALL blasts are more resistant than

those from children to several drugs used in ALL treatment, suggesting that different mechanism can contribute to or modulate constitutive and acquired resistance. Adding to this is the versatile nature of MRP4, reflected in affinity for a variety of substrates that can, upon exposure, differentially regulate MRP4 expression.[4]

There are several differences in treatment modalities between childhood ALL DFCI protocol and GMALL adult ALL trial regarding drug type and dose, and schedule of administration.[5,6] Beside noted difference in MTX dose, high-dose MTX in GMALL trial is given at several instances during consolidation phase compared with earlier single administration during remission induction in DFCI protocol.

The association of *MRP4* polymorphisms seen in childhood ALL seems to be more apparent in the standard risk group, as reflected in our article's data supplement.[1] Brüggemann et al report the analysis of standard risk patients only. However, the risk classes between 2 protocols are not comparable. Beside already mentioned age difference, the GMALL protocol does not encounter CNS disease at presentation among risk classification criteria, whereas the standard risk groups also includes the subset of T-cell leukemia.[5,6]

One possible explanation for association found in our study[1] is that higher frequency of toxicity in A934 carriers would lead to more frequent drug withdrawal or dose reduction, which might cause higher frequency of relapse. However, this reduction probably would not be sufficient to explain reduction in event-free survival, and other mechanisms contribute as well. For instance, MRP4 participates also in efflux of folate; down-regulation of MRPs might result in decreased folate efflux, thereby leading to expansion of the intracellular folate pool and antifolate resistance.[7] This further illustrates the complexity of MRP effects and regulation.

In conclusion, we believe that the report of Brüggemann et al is important, as it highlights the applicability of pharmacogenetic findings in childhood ALL to adults, yet further studies analyzing more comparable childhood ALL populations are needed to establish the role of *MRP4* polymorphisms.

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**Conflict-of-interest disclosure:** The authors declare no competing financial interests.

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## References

1. Ansari M, Sauty G, Labuda M, et al. Polymorphisms in multidrug resistance-associated protein gene 4 is associated with outcome in childhood acute lymphoblastic leukemia. *Blood.* 2009;114:1383-1386.
2. Ansari M, Krajinovic M. Pharmacogenomics of acute leukemia. *Pharmacogenomics.* 2007;8:817-834.
3. Plasschaert SL, Kamps WA, Vellenga E, et al Prognosis in childhood and adult acute lymphoblastic leukaemia: a question of maturation? *Cancer Treat Rev.* 2004;30:37-51.
4. Russel FG, Koenderink JB, Masereeuw R. Multidrug resistance protein 4 (MRP4/ABCC4): a versatile efflux transporter for drugs and signalling molecules. *Trends Pharmacol Sci.* 2008;29:200-207.
5. Moghrabi A, Levy DE, Asselin B, et al. Results of the Dana-Farber Cancer Institute ALL Consortium Protocol 95-01 for children with acute lymphoblastic leukemia. *Blood.* 2007;109:896-890.
6. Brüggemann M, Raff T, Flohr T, et al. Clinical significance of minimal residual disease quantification in adult patients with standard-risk acute lymphoblastic leukemia. *Blood.* 2006;107:1116-1123.
7. Assaraf YG. The Role of multudrug resistance efflux transporters in antifolate resistance and folate homeostasis. *Drug Resist Updat.* 2006;9:227-246.

# To the editor:

## Central nervous system prophylaxis in mantle cell lymphoma

In their thorough review of mantle cell lymphoma (MCL),[1] Ghielmini and Zucca describe that central nervous system (CNS) disease involvement, albeit rare at presentation, has an incidence of 4% to 22% in relapsed patients. Nevertheless, they do not state their recommendations regarding prophylaxis to MCL patients. The National Comprehensive Cancer Network (www.nccn.org) guidelines for diffuse large B-cell (DLBCL) lymphoma recommend CNS prophylaxis with 4 to 8 doses of intrathecal methotrexate (MTX) and/or cytarabine for patients with aggressive lymphomas who have paranasal sinus, testicular, epidural, bone marrow, 2 extranodal site involvement, or HIV lymphomas. The guidelines for MCL patients further suggest lumbar puncture at diagnosis for patients with blastic variants or with neurologic symptoms, but do not state any recommendations for prophylactic treatment at either diagnosis or relapse. Because there are no definite guidelines, and most patients are not treated with CNS-penetrating agents such as high dose MTX/cytarabine regimens,[1] it is left to the physician's discretion whether to administer CNS prophylaxis or not. For instance, in the United Kingdom, one-third of hematologists administer prophylaxis to MCL patients and only in specific circumstances.[2] Among the many reports of MCL in the literature, 4 concentrated their efforts on direct assessment of CNS involvement in MCL,[3-6] and all are retrospective (Table 1); therefore, the true incidence is unknown.

Interestingly, although most patients with CNS involvement develop symptoms at or after first relapse, not only the blastoid variant MCL had this propensity while having a higher risk. It might be argued that since most patients will succumb to their disease, which is rarely curable, and because CNS disease is only one part of a systemic relapse,[3] there is limited application for primary prophylaxis. Nevertheless, in all reports, CNS disease was associated with worse overall survival. In addition, with the advent of newer agents and curative treatment attempts,