Overexpression of survivin and its variants in the bone marrow of patients with chronic myeloid leukaemia (CML) in remission: Restricting their use in immunotherapy?

Speletas M,[1] Gramoustianou E-S,[1] Loules G,[1] Tsochas S,[1] Kalala F,[1] Braimi M,[2] Mandala E,[3] Katodritou E,[4] Karanikas V,[1] Germenis AE[1]

[1]Department of Immunology and Histocompatibility, School of Medicine, University of Thessaly, University Hospital of Larissa, GR-411 10 Larissa, Greece; [2]Hematology Department, Papageorgiou Hospital, Thessaloniki, [3]4th Department of Internal Medicine, Aristotle University of Thessaloniki, [4]Hematology Department, Theagenion Anticancer Hospital, Thessaloniki

Survivin is an inhibitor of apoptosis, expressed during development and overexpressed in human cancers. In CML, its expression has been attributed to the activation of chimeric bcr-abl protein pathways, conferring growth advantage to neoplastic cells and playing a critical role in leukemogenesis. Survivin specific cytotoxic T-cell clones have been determined in CML, implying that immunotherapy against survivin may be an alternative therapeutic approach for patients not achieving an hematologic, cytogenetic or molecular remission.[1-3] The aim of this study was to analyse the expression of survivin and its variants (pro- and anti-apoptotic) in CML patients receiving imatinib both at diagnosis and during the follow-up period.

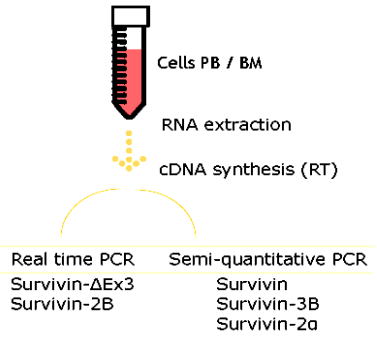
Materials

✓ 20 patients with CML ✓ 12 normal volunteers

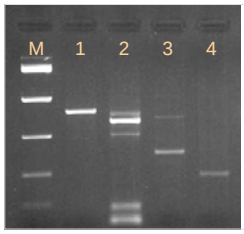
(normal individuals and patients with non-Hodgkin lymphoma in complete remission)

Methods

Expression of survivin and its variants



RT-PCR (Real-Time and Semi-quantitative)



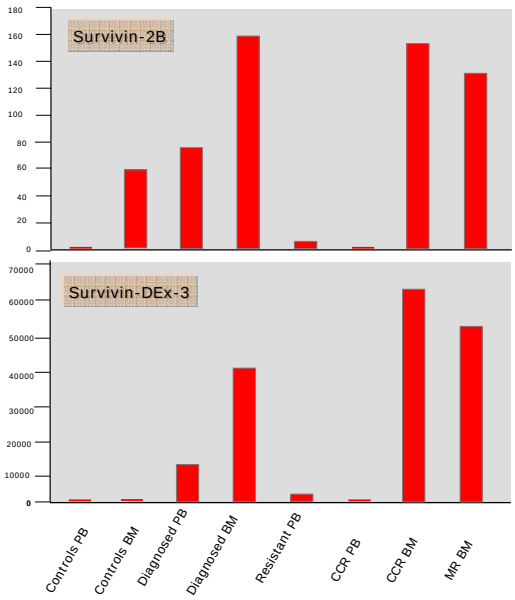
1: b-actin
2: STD survivin
3: survivin-2a
4: survivin-3B

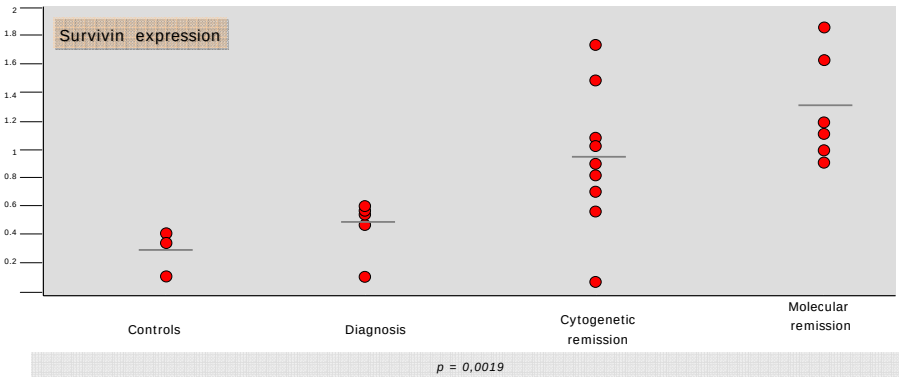
Gene	Forward Primer	Reverse Primer
REAL-TIME PCR		
Survivin-2B	*CUO50* 5' TTCTCAAGGACCACCGCATC 3'	*CUO51* 5' TGTTCCTCTCTCGTGATCCG 3'
Survivin-ΔEx3	*CUO52* 5' CTCAAGGACCACCGCATCTCTAC 3'	*CUO53* 5' GGTTTCCTTTGCATGGGGTCGTC 3'
CONVENTIONAL PCR		
Survivin-STD	*HV53F* 5' CAGATTTGAATCGCGGGACCCGTT 3'	*HV54R* 5' CCTGGAAGTGGTGCAGCCACTCTG 3'
Survivin-3B	*CUO54* 5' GGTTGCGCTTTCCTTTCTGTC 3'	*CUO55* 5' TGGCAAACATCAGGCTCTTCC 3'
Survivin-2α	*CUO56* 5' TCAAGGACCACCGCATCTCT 3'	*CUO57* 5' AGCAATGAGGGTGGAAAGCA 3'

Primers used in real-time and semi-quantitative PCR reactions. B-actin and b2M (b2-microglobulin) were used as references genes in the conventional and quantitative real time PCRs, respectively.

The disease burden (bcr-abl transcripts) was estimated by standard protocol (quantitative real-time RT-PCR, Ipsogen, UK). REST and SPSS-v.10 software were used for the statistical analysis.

Results





Increased expression of survivin and its variants was observed in all CML patients at diagnosis, both in PB (peripheral blood) and BM (bone marrow). However, such an increased expression was also found in BM of CML patients in complete cytogenetic and/or molecular remission of the disease, which was considerably higher than its expression at diagnosis or in normal BM.

Conclusion

The increased expression of survivin in BM of CML patients during remission could be attributed to the presence of normal progenitor cells that rehabilitate a normal haematopoiesis after treatment. These findings question the suitability of survivin as a target in immunotherapy target in CML patients as it was previously proposed.

References

1. Carter et al. Regulation of survivin expression through Bcr-Abl/MAPK cascade: targeting survivin overcomes imatinib resistance and increases imatinib sensitivity in imatinib-responsive CML cells. Blood 2006;107:1555-63.
2. Hernández-Boluda et al. Survivin expression in the progression of chronic myeloid leukemia: a sequential study in 16 patients. Leuk Lymphoma 2005;46:717-22.
3. Conte et al. Survivin expression in chronic myeloid leukemia. Cancer Lett 2005;225:105-10.

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