rituximab, can kill CLL cells via the mitochondrial apoptotic pathway. Increased BCL-X_L and BFL-1/A1 would therefore be expected to cause resistance to apoptosis induced by these conventional agents. These results suggest that strategies to drive CLL cells from their lymph node niches might be an important prerequisite for killing them, not only by BCL-2 antagonists, but by conventional agents as well. Little is known about the equilibrium between the circulating state and the lymph node resident state in CLL, but the results of Vogler et al suggest that this may be a critically important area of study to improve CLL treatment.

Conflict-of-interest disclosure: A.L. is co-founder of Eutropics Pharmaceuticals.

REFERENCES

1. Del Gaizo Moore V, Brown JR, Certo M, et al. Chronic lymphocytic leukemia requires BCL2 to sequester prodeath BIM, explaining sensitivity to BCL2 antagonist ABT-737. J Clin Invest. 2007;117:112-121.
2. Oltersdorf T, Elmore SW, Shoemaker AR, et al. An inhibitor of Bcl-2 family proteins induces regression of solid tumours. Nature. 2005;435:677-681.
3. Vogler M, Butterworth M, Majid A, et al. Concurrent up-regulation of BCL-X_L and BCL2A1 induces approximately 1000-fold resistance to ABT-737 in chronic lymphocytic leukemia. Blood. 2009;113:4403-4413.
4. Munk Pedersen I, Reed J. Microenvironmental interactions and survival of CLL B-cells. Leuk Lymphoma. 2004;45:2365-2372.
5. Willimott S, Baou M, Naresh K, et al. CD154 induces a switch in pro-survival Bcl-2 family members in chronic lymphocytic leukaemia. Br J Haematol. 2007;138:721-732.

LYMPHOID NEOPLASIA

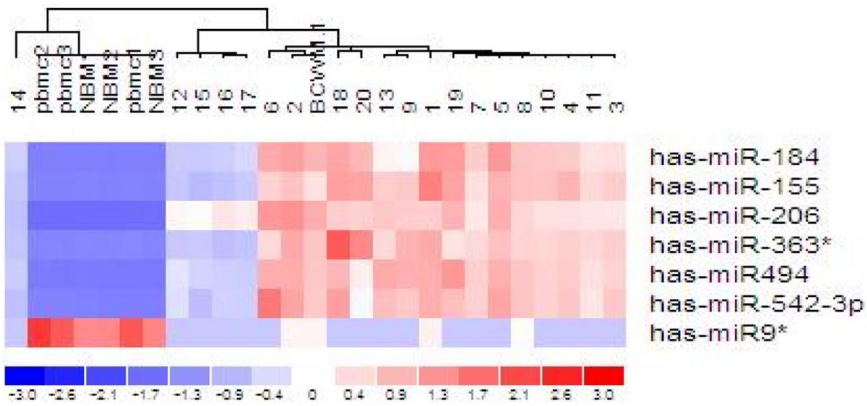
Comment on Roccaro et al, page 4391

MicroRNAs to know in Waldenström macroglobulinemia

Angelo Vacca and Franco Dammacco UNIVERSITY OF BARI MEDICAL SCHOOL

In this issue of *Blood*, Roccaro and colleagues evaluate the crucial role of miRNAs in regulating the biology and prognosis of Waldenström macroglobulinemia, providing in vitro and in vivo evidences for miRNA-based targeted therapies in this disease.

Epigenetic modifications at the level of microRNAs (miRNAs) have recently gained considerable attention in the field of cancer research. The miRNAs are short, noncoding RNAs that negatively regulate gene expression by binding to the 3′ untranslated region of the target mRNAs, leading to mRNA degradation or inhibition of translation.[1,2] They have been described as playing crucial roles in regulating physiological processes as well as tumor pathogenesis. Indeed, much evidence has clearly demonstrated that miRNA expression profiles differ between normal and tumor tissues, both in solid and hematologic malignancies.[3-6]



Supervised hierarchical clustering analysis demonstrates differential expression of miRNA patterns in WM patients(1-20) as compared with healthy subjects (NBM1-3; pbmc1-3), shown by the intensity of red (up-regulation) versus blue (down-regulation). See the complete figure in the article beginning on page 4391.

In this issue, Roccaro et al evaluate for the first time the miRNA signature in Waldenström macroglobulinemia (WM).[7] They identify increased expression of miRNAs-*363**, *-206*, *-494*, *-155*, *-184*, *-542–3p*, and decreased expression of miRNA-*9** in primary bone marrow–derived WM tumor cells. Based on this first observation, they wondered next whether the miRNA signature could be linked to prognosis in these patients, and how miRNAs could functionally contribute to WM pathogenesis. The authors show that increased expression of the 6 miRNAs significantly correlated with a poorer outcome as predicted by the International Prognostic Staging System. Moreover, in vitro and in vivo studies clearly demonstrated that, among those deregulated miRNAs, miRNA-*155* is likely involved in WM biology. The studies showed that miRNA-*155* specifically targets WM cells even in the context of a bone marrow milieu by inhibiting MAPK/ERK, PI3/AKT, and NF-*k*B pathways, which are known to be constitutively activated in WM as well as in other B-cell malignancies.[8]

The importance of the findings put forth by Roccaro et al is substantial. If cytogenetic and molecular studies on gene expression analysis at the miRNA level have demonstrated minimal changes in WM cells,[9] the described significant differences in WM miRNA expression profiling improve our understanding of the underlying molecular changes that lead to the initiation and progression of this rare disease. Also, miRNA-*155* may be regarded as a sufficiently restricted therapeutic target in refractory-resistant WM.

These studies raise a few questions. It is well known that WM represents a rare B-cell malignancy, with an incidence of 3 cases per 1 000 000 persons each year, accounting for approximately 1% to 2% of all hematologic malignancies. Roccaro et al have collected and studied 20 primary bone marrow WM samples, and they did not observe differences after supervised clustering analysis between untreated and treated patients, indicating that samples had similar expression patterns. It would thus be interesting to enlarge the 2 cohorts of samples in further studies to see if any patterns correspond to disease relapse or progression. In addition, while the authors showed that miRNA-*155* negatively regulates the canonical NF-*k*B pathway, it would be worthwhile to understand the possible effects of miRNA-*155* on the noncanonical NF-*k*B pathway as well.

In conclusion, these innovative and very well conducted studies represent a major achievement in the field of hematologic malignancies. They bring miRNAs nearly from the bench to the bedside because they provide the preclinical evidence for the development of novel miRNA-based prognostic and therapeutic options in WM.

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

REFERENCES

1. Xie X, Lu J, Kulbokas EJ, et al. Systematic discovery of regulatory motifs in human promoters and 3′UTRs by comparison of several mammals. Nature. 2005;434:338-345.
2. He L, Hannon GJ. MicroRNAs: small RNAs with a big role in gene regulation. Nat Rev Genet. 2004;5:522-531.
3. Roldo C, Missiaglia E, Hagan JP, et al. MicroRNA expression abnormalities in pancreatic endocrine and acinar tumors are associated with distinctive pathologic features and clinical behaviour. J Clin Oncol. 2006;24:4677-4684.
4. Calin GA, Liu CG, Sevignani C, et al. MicroRNA profiling reveals distinctive signatures in B cell chronic lymphocytic leukemias. Proc Natl Acad Sci U S A. 2004;101:11755–11760.
5. Eis PS, Tam W, Sun L, et al. Accumulation of miR-155 and BIC RNA in human B cell lymphomas. Proc Natl Acad Sci U S A. 2005;102:3627-2632.
6. Rai D, Karanti S, Jung I, Dahia PL, Aguiar RC. Coordinated expression of microRNA-155 and predicted target genes in diffuse large B-cell lymphoma. Cancer Genet Cytogenet. 2008;181:8-15.
7. Roccaro AM, Sacco A, Chen C, et al. microRNA expression in the biology, prognosis, and therapy of Waldenstrom macroglobulinemia. Blood. 2009;113:4391-4402.
8. Leleu X, Eeckhoute J, Jia X, et al. Targeting NF-kappaB in Waldenström macroglobulinemia. Blood. 2008;111:5068-5077.
9. Gutiérrez NC, Ocio EM, de Las Rivas J, et al. Gene expression profiling of B lymphocytes and plasma cells from Waldenström's macroglobulinemia: comparison with expression patterns of the same cell counterparts from chronic lymphocytic leukemia, multiple myeloma and normal individuals. Leukemia. 2007;21:541-549.

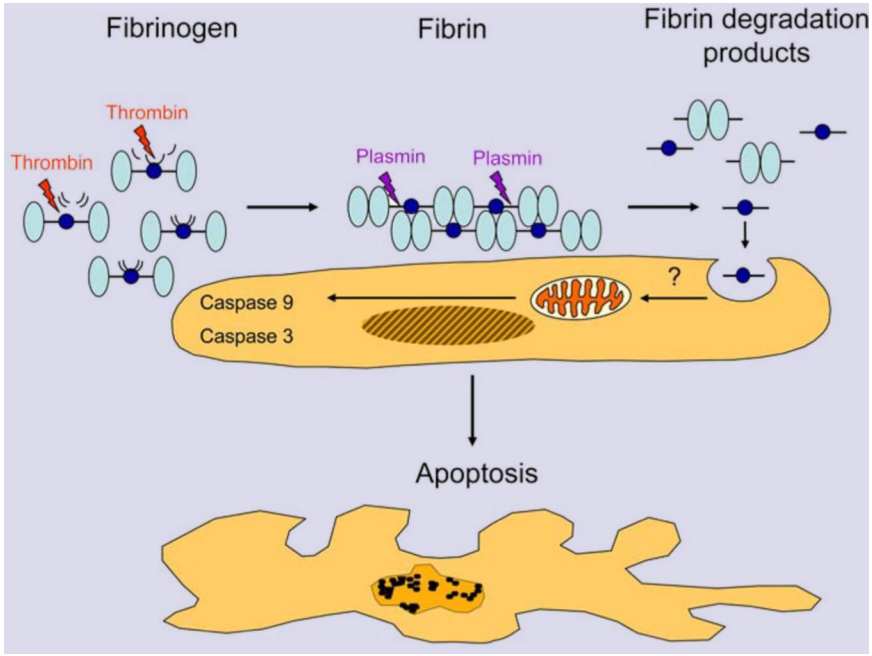
● ● ● THROMBOSIS & HEMOSTASIS

Comment on Guo et al, page 4431

How can fibrinolysis induce cell death?

Margarethe Geiger MEDICAL UNIVERSITY OF VIENNA

In this issue of *Blood*, Guo and colleagues elucidate molecular details of a novel mechanism, linking the degradation of fibrin (but not fibrinogen) by plasmin to apoptosis of placental trophoblast cells.



Fibrin is formed by the action of thrombin on fibrinogen releasing fibrinopeptides A and B from the Aα- and Bβ-chains of fibrinogen. Plasmin generated by the plasminogen activators uPA or tPA cleaves fibrin into fibrin degradation products (shown here: D-dimer, light blue, and fragment E, dark blue). Fibrin fragment E is internalized by cells and induces apoptosis of trophoblast cells. Illustration by Thomas Nardelli.

There is convincing evidence from the literature that disorders of the maternal coagulation system are associated with complications of pregnancy including fetal loss. In many studies, an association between fibrin deposition in the placenta and apoptosis of trophoblast cells has been observed.[1] The group of Weiler and collaborators have shown previously that mouse embryos deficient in the thrombin receptor thrombomodulin (*Thbd*$^{-/-}$) die before the development of a functional cardiovascular system because of a defect in the placenta.[2] In a more recent study that analyzed the mechanisms responsible for the placental defect in these mice, they found that activated coagulation factors induce growth inhibition and apoptosis of placental trophoblast cells.[3] While the growth-inhibiting effect can be attributed to the activation of protease-activated receptors, cell death is caused by degradation products of fibrin. Neither fibrinogen nor intact fibrin nor degradation products of fibrinogen caused apoptosis.

In this issue of *Blood*, Guo et al perform a study based on these previous findings that discloses the structural requirements and molecular mechanisms involved in fibrin degradation product-mediated cell death.[4] They show that the apoptosis-inducing activity is not restricted to the mouse trophoblast system but is also seen with human fibrin degradation products and a variety of cell types. They can furthermore assign the apoptosis-inducing activity to a sequence within the Aα-chain of fibrin fragment E, which has to be cleaved by thrombin as well as by plasmin to gain apoptosis-inducing activity. Part of the pro-apoptotic activity can be attributed to an RGD-motif, but the majority is RGD-independent. Induction of apoptosis by fibrin fragment E requires its uptake by the cell. Uptake, but not apoptosis, is mediated via a motif located within the sequence Aα52-81. Apoptosis-inducing activity itself is located in Aα17-37. The internalization of fibrin fragment E, but not the intracellular mechanism mediating apoptosis, is caveolin-1–dependent. Intracellular pathways have not yet been analyzed in detail, but data presented suggest activation of the mitochondrial pathway and involvement of caspases 9 and 3.

This newly described pathway linking fibrin degradation to apoptosis may play a role in several physiologic and pathophysiologic situations. It may lead to trophoblast cell death, causing placental insufficiency and