REFERENCES

Aifantis, I., Raetz, E., and Buonamici, S. (2008). Nat. Rev. Immunol. *8*, 380–390.

Burkhardt, B. (2010). Br. J. Haematol. *149*, 653–668.

Campana, D. (2010). Curr. Hematol. Malig. Rep. *5*, 169–176.

Coustan-Smith, E., Mullighan, C.G., Onciu, M., Behm, F.G., Raimondi, S.C., Pei, D., Cheng, C., Su, X., Rubnitz, J.E., Basso, G., et al. (2009). Lancet Oncol. *10*, 147–156.

Feng, H., Stachura, D.L., White, R.M., Gutierrez, A., Zhang, L., Sanda, T., Jette, C.A., Testa, J.R., Neuberg, D.S., Langenau, D.M., et al. (2010). Cancer Cell *18*, this issue, 353–366.

Karrman, K., Forestier, E., Heyman, M., Andersen, M.K., Autio, K., Blennow, E., Borgstrom, G., Ehrencrona, H., Golovleva, I., Heim, S., et al. (2009). Genes Chromosomes Cancer *48*, 795–805.

Mullighan, C.G., Su, X., Zhang, J., Radtke, I., Phillips, L.A., Miller, C.B., Ma, J., Liu, W., Cheng, C., Schulman, B.A., et al. (2009). N. Engl. J. Med. *360*, 470–480.

Palomero, T., Lim, W.K., Odom, D.T., Sulis, M.L., Real, P.J., Margolin, A., Barnes, K.C., O'Neil, J., Neuberg, D., Weng, A.P., et al. (2006). Proc. Natl. Acad. Sci. USA *103*, 18261–18266.

Raetz, E.A., Perkins, S.L., Bhojwani, D., Smock, K., Philip, M., Carroll, W.L., and Min, D.J. (2006). Pediatr. Blood Cancer *47*, 130–140.

Weng, A.P., Ferrando, A.A., Lee, W., Morris, J.P., IV, Silverman, L.B., Sanchez-Irizarry, C., Blacklow, S.C., Look, A.T., and Aster, J.C. (2004). Science *306*, 269–271.

Epigenetics and MicroRNAs Combine to Modulate the MDM2/p53 Axis in Myeloma

Marta Chesi[1] and P. Leif Bergsagel[1,*]
[1]Division of Oncology/Hematology, Mayo Clinic in Arizona, Scottsdale AZ, 85259, USA
*Correspondence: bergsagel.leif@mayo.edu
DOI 10.1016/j.ccr.2010.10.004

Avoiding the inhibitory effects of p53 on cell growth is important for tumor progression. In this issue of *Cancer Cell*, Picchiorri et al. describe epigenetic silencing of MDM2-targeting microRNAs in multiple myeloma (MM), which generally appears to have intact p53 function. This provides the rationale for microRNA-targeted therapy for MM.

Genetically, multiple myeloma (MM) can be divided into tumors with recurrent immunoglobulin heavy chain gene translocations or hyperdiploidy. Superimposed on this are a variety of presumably secondary genetic events, including activating mutations of *Ras*, rearrangements of *Myc*, mutations that activate the NFkB pathway, and late inactivating mutations of p53 (Fonseca et al., 2009). As noted by Pichiorri et al., (2010), mutations of p53 are rare in untreated MM and the tumors appear to have intact, if perhaps suppressed, p53 function, suggesting that therapeutic modulation of the p53/MDM2 pathway holds promise to help the majority of patients.

Deletion of one copy of p53 by FISH has been uniformly found to be an adverse prognostic factor with all therapies used in the treatment of MM: alkylating agents, proteasome inhibitors, and immune modulator-based therapies (Lode' et al., 2010; Kapoor et al., 2010). In contrast to patients with other poor-prognostic genetic lesions [e.g., t(4;14)], patients with deletion of p53 have seen less improvement in their survival with the addition of proteasome inhibitors and immune modulators. Nevertheless, data concerning loss of heterozygosity of p53 due to deletion of chromosome 17p and/or p53 mutation in MM have been confusing, perhaps at least in part because of old literature based on the analysis of unpurified bone marrow samples. In an analysis of CD138-purified myeloma cells from 716 patients, deletion of one copy of p53 was detected by FISH in 3% of MGUS (monoclonal gammopathy of undetermined significance), 1% of SMM (smoldering multiple myeloma), and 10% of MM (Chiecchio et al., 2009). There is a paucity of data on patients at the time of relapse; however, in another recent study, deletion of p53 was seen in 56% of primary plasma cell leukemia and 83% of the more aggressive secondary plasma cell leukemia (Tiedemann et al., 2008).

Somewhat surprising, given the strong correlation between loss of one copy of p53 deletion and survival, is the fact that in a cohort of 92 untreated MM patients, only 37% of patients with p53 deletions by FISH were found to have mutations on the remaining allele, although all those with mutations were found to also have p53 deletions (Lode' et al., 2010). It would be very informative to characterize whether in terminal samples from patients that initially presented with a 17p deletion by FISH, a p53 mutation has emerged under the selective pressure of therapy and disease progression. In the absence of such a study, we can examine the status of p53 in a panel of human MM cell lines derived from patients at the very end stages of the disease. It is notable that the majority of MM cell lines have p53 mutation (Mazars et al., 1992). Although one has to consider that cell lines have been under the selective pressure of being

able to grow ex vivo, these data strongly argue that is the ultimate inactivation of p53 that underlies the poor prognosis associated with the initial loss of one copy.

What is clear from all these studies and in the absence of more definitive sequencing data is that the great majority of patients seems to retain an intact p53 pathway and may therefore be susceptible to its successful modulation. One well-characterized way by which p53 is modulated is the E3 ubiquitin ligase MDM2, and more recent studies have identified that microRNAs 29 and 125a target p53 and members of its pathway (Park et al., 2009; Zhang et al., 2009). In order to determine the pathways relevant to MM, Pichiorri et al. start their analysis by identifying in MM cell lines the p53-regulated miRNAs miR-192, 194, and 215 (Pichiorri et al., 2010). Key to these studies is a panel of genetically characterized MM cell lines with and without inactivating mutations of p53. They determine that these miRs are direct transcriptional targets of p53, and, intriguingly, they find that the miRs are expressed at a lower level in MM than MGUS and are associated with promoter hypermethylation in MM cell lines (Figure 1). They show that expression of these miRs induces a cell cycle arrest in a p53-dependent manner and, noting lower levels of MDM2, they go on to show that these miRs directly target MDM2, with the expected downstream effects on IGF1 and IGF1R migration and invasion.

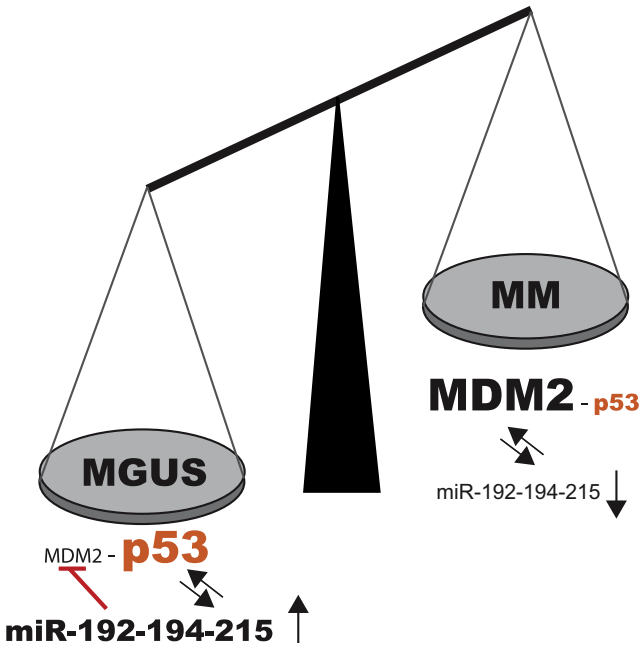

Figure 1. Illustration of the p53–miR-*192,194,215*–MDM2 Auto Regulatory Loops
An illustration of the p53–miR-*192,194,215*–MDM2 auto regulatory loops showing the central role played by the miRs in determining the balance of p53 suppressor and the MDM2 oncoprotein expression levels (adapted from Pichiorri et al., 2010).

So how close are we to miR replacement therapy for MM? The authors do not conjecture, and they leave a few other questions unanswered. They postulate, but do not demonstrate, that aberrant promoter methylation is responsible for silencing of these miRs during the transition from MGUS to MM. They suggest a gradient effect of p53 function that in patients with 17p deletion is modulated by haploinsuffiency. During tumor progression, due to a partial loss of expression of regulatory miRs, presumably this is sufficient to allow intramedullary tumor growth in vivo but, as noted above, is presumably insufficient to allow in vitro tumor growth as MM cell lines. These studies provide the rationale for the development of therapies to supplement inappropriately suppressed miR expression in MM.

REFERENCES

Chiecchio, L., Dagrada, G.P., Ibrahim, A.H., Dachs Cabanas, E., Protheroe, R.K., Stockley, D.M., Orchard, K.H., Cross, N.C., Harrison, C.J., and Ross, F.M.; UK Myeloma Forum. (2009). Haematologica *94*, 1708–1713.

Fonseca, R., Bergsagel, P.L., Drach, J., Shaughnessy, J., Gutierrez, N., Stewart, A.K., Morgan, G., Van Ness, B., Chesi, M., Minvielle, S., et al; International Myeloma Working Group. (2009). Leukemia *23*, 2210–2221.

Kapoor, P., Fonseca, R., Rajkumar, S.V., Sinha, S., Gertz, M.A., Stewart, A.K., Bergsagel, P.L., Lacy, M.Q., Dingli, D.D., Ketterling, R.P., et al. (2010). Mayo Clin. Proc. *85*, 532–537.

Lode', L., Eveillard, M., Trichet, V., Soussi, T., Wuilleme, S., Richebourg, S., Magrangeas, F., Ifrah, N., Campion, L., Traulle, C., et al. (2010). Haematologica, in press. Published online July 15, 2010. 10.3324/haematol.2010.023697.

Mazars, G.R., Portier, M., Zhang, X.G., Jourdan, M., Bataille, R., Theillet, C., and Klein, B. (1992). Oncogene *7*, 1015–1018.

Park, S.Y., Lee, J.H., Ha, M., Nam, J.W., and Kim, V.N. (2009). Nat. Struct. Mol. Biol. *16*, 23–29.

Pichiorri, F., Sung-Suk, S., Rocci, A., De Luca, L., Taccioli, C., Santhanam, R., Wenchao, Z., Benson, D.M., Jr., Hofmainster, C., Alder, H., et al. (2010). Cancer Cell *18*, this issue, 367–381.

Tiedemann, R.E., Gonzalez-Paz, N., Kyle, R.A., Santana-Davila, R., Price-Troska, T., Van Wier, S.A., Chng, W.J., Ketterling, R.P., Gertz, M.A., Henderson, K., et al. (2008). Leukemia *22*, 1044–1052.

Zhang, Y., Gao, J.S., Tang, X., Tucker, L.D., Quesenberry, P., Rigoutsos, I., and Ramratnam, B. (2009). FEBS Lett. *583*, 3725–3730.