

## Commentary: Involvement of H-ras in Erythroid Differentiation of TF1 and Human Umbilical Cord Blood CD34<sup>+++</sup> Cells

I read with interest this paper by Ge Y *et al.*, describing the consequences of retrovirally mediated transfer of the H-*ras* gene into a human erythroleukemia cell line TF-1 (1). The authors find elevated levels of pre-mRNA for the erythropoietin receptor (EpoR), as well as a slight increase in mRNA for  $\beta$ - and  $\gamma$ - globins. Levels of mRNA for EpoR, however, were not upregulated; also, GATA-1 mRNA levels did not change. These data, while intriguing, raise the question of whether or not *ras* plays a role in regulating erythroid differentiation.

Winkelman JC *et al.* had previously identified a translocation breakpoint in exon 8 of the EpoR gene in TF-1 cells (2). Most of the EpoR mRNA in TF-1 cells was produced by this translocated gene, whereas the normal EpoR gene was expressed only minimally (2). It is thus possible that in this cell line, the elevated expression of the 4 Kb pre-mRNA could be a result of deregulated transcriptional control of the translocated gene, caused by the exogenously introduced *ras* gene.

Most hematopoietic growth factors stimulate *Ras* and the *Ras*/mitogen-activated protein (MAP) kinase pathway in cultured cell systems. Activating mutations of *Ras* occur in 25-50% of acute myeloid leukemia and myelodysplastic syndromes (3,4), and the *Ras* signal transduction pathway is presumed to be activated in many other types of human cancers. However, there exists no direct link between the induction of differentiation and the activation state of *Ras* or of the *Ras*/MAP kinase pathway in hematopoietic system. Instead, many data show that *ras* activation constitutes part of the signals in the mitogenic pathway and not necessarily the differentiation pathway, triggered by various hematopoietic factors. The importance of *ras* gene is suggested by its involvement in the signal transduction for mitogenic responses activated by Epo (5), IL-3, GM-CSF, stem cell factor, IL-2, IL-5 (6), and M-CSF (7). Data shown in Figure 5A of

Ge Y *et al.*'s paper also suggest that there was a significant increase in the clonogenic potential among colonies for HPP-CFC/CFU-GM, as well as BFU-E/CFU-GEMM, of the umbilical cord blood CD34<sup>+++</sup> cells transduced with H-*ras*. [It is not clear whether there was also an increase in the size of these colonies, as shown for TF-1 cells in Figure 4B of the paper (1).] Before *ras* may be assigned a role in differentiation, it must first be demonstrated that the *Ras*/MAP kinase pathway is in fact involved in differentiation *per se* and not proliferation.

### REFERENCES

1. Ge Y, Li Z, Marshall MS, Broxmeyer HE, Lu L. Involvement of H-ras in Erythroid Differentiation of TF1 and Human Umbilical Cord Blood CD34<sup>+++</sup> Cells. *Blood Cells Mol Dis* 24:124-136, 1998.
2. Winkelman JC, Ward JC, Mayeux P, Lacombe C, Schimmenti L, Jenkins, RB. A translocated erythropoietin receptor gene in a human erythroleukemia cell line (TF-1) expresses an abnormal transcript and a truncated protein. *Blood* 85:179-185, 1995.
3. Lubbert, M, Mirro J. Jr., Kitchingman, G., McCormick, F., Mertelsmann, R., Herrmann, F., Koeffler, HP. Prevalence of N-*ras* mutations in children with myelodysplastic syndromes and acute myeloid leukemia. *Oncogene* 7, 263-268, 1992.
4. Sawyers, CL, McLaughlin, J, Witte, ON. Genetic requirement for Ras in the transformation of fibroblasts and hematopoietic cells by the Bcr-Abl oncogene. *J Exp Med* 181, 307-313, 1995.
5. Torti M, Marti KB, Altschuler D, Yamamoto K, Lapetina EG. Erythropoietin induces p21*ras* activation and p120GAP tyrosine phosphorylation in human erythroleukemia cells. *J Biol Chem* 267:8293-8298, 1992.
6. Duronio V, Welham MJ, Abraham S, Dryden P, Schrader JW. p21 *ras* activation via hemopoietin receptors and c-kit requires tyrosine kinase activity but not tyrosine phosphorylation of p21*ras* GTPase-activating protein. *Proc Natl Acad Sci U S A* 89:1587-1591, 1992.
7. Bortner DM, Ulivi M, Roussel MF, Ostrowski MC. The carboxy-terminal catalytic domain of the GTPase-activating protein inhibits nuclear signal transduction and morphological transformation mediated by the CSF-1 receptor. *Genes Dev* 5:1777-1785, 1991.

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