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Thioredoxin redox regulation in p53 activity

Redox regulation is critical for the functioning of a number of transcription factors. The p53 protein has the structural and biochemical properties of a redox-regulated transcription factor, but the role of redox regulation in the control of p53 activity is rather poorly understood. Thioredoxin (Trx) is a small, ubiquitous, multifunctional, stress-inducible protein with a redox-active disulfide/dithiol within its active site sequence. Trx operates together with Trx reductase (TRR), an NADPH-dependent FAD-containing enzyme, as a protein disulfide reducing system. Yeast deficient for TRR accumulate p53 protein in a transcriptionally inactive form. In mammalian cells, p53 binds to redox factor-1 (APE/Ref-1), an enzyme containing an abasic endonuclease domain involved in base excision repair (BER). APE/Ref-1 also has a thiol reductase domain recycled by Trx, involved in regulating AP-1.

In a recent study (Seemann and Hainaut¹), the involvement of TRR and APE/Ref-1 in the control of p53 protein activity was examined. They found that silencing of TRR and modulation of intracellular distribution of Trx contributed to enhanced p53 protein activation by increasing basal p53 levels and by facilitation of its accumulation into the nucleus. The modulation was exerted by oxidized, rather than reduced Trx. APE/Ref-1 was required as an intermediate for oxidized Trx to modulate p53 activity in DNA binding, nuclear translocation and protein stabilization. Inhibition of APE/Ref-1 accelerated p53 protein turnover, resulting in a decrease in p53 levels and activity. When both factors (TRR and APE/Ref-1) were inhibited, basal levels of p53 were restored. Inhibition of either factor, however, did not prevent activation and accumulation of p53 in response to DNA damage by doxorubicin.

The findings in this study suggest that the interaction between p53, APE/Ref-1 and Trx may actually serve as a critical crossroad between redox sensing (through Trx), BER (through APE/Ref-1), and DNA-damage sensing (through p53). Furthermore, the redox sensitivity of p53 may provide connection between p53 activity and the sensing and repair of DNA damage.

In this issue of *Laboratory Investigation*, Kawasaki *et al*² studied the roles of Trx in the host defense mechanism against *Helicobacter felis* infection. They found that Trx suppressed oxidative cytotoxicity and DNA damage, and inhibited neutrophil migration both *in vivo* and *in vitro*. Both studies provide

insights to oxidative cytotoxicity and its effect in DNA-damage repair and sensing.

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References

- 1 Seemann S, Hainaut P. Roles of thioredoxin reductase 1 and APE/Ref-1 in the control of basal p53 stability and activity. *Oncogene* 2005;24: 3853–3863.
- 2 Kawasaki K, Nishio A, Nakamura H, *et al*. *Helicobacter felis*-induced gastritis was suppressed in mice overexpressing thioredoxin-1. *Lab Invest* 2005;85:1104–1117.

Genetic differences between primary colorectal carcinomas and their metastases

Colorectal carcinoma is one of the most frequent malignancies in the industrialized world, with a relatively high mortality rate mostly due to metastatic spread of the disease. Substantive insights have been made into the multistep molecular pathogenesis that transforms normal colonic epithelium into a primary colorectal carcinoma. However, the genetic mechanisms that enable a primary colorectal carcinoma to transform into a metastatic phenotype have not been as extensively studied. Comparative genomic hybridization (CGH) is a new and powerful technique that can evaluate the entire tumor genome for chromosomal gains or losses without the need for cell cultures. These chromosomal gains or losses are likely to contain relevant information on the genes that are involved in malignant transformation.

In an attempt to better understand the genetic underpinnings of pulmonary metastases from colorectal carcinoma, Jiang *et al*¹ used CGH to compare the genetic profiles of 18 paired samples. The techniques involved purifying the paraffin-embedded tumor and reference DNA, which were respectively labeled with Green and Red-dUTP, and combining this with unlabeled human Cot-1 DNA. The resulting probe mixture was denatured, placed onto a metaphase chromosome slide and hybridized for 3 days. The chromosomes were counterstained and the fluorescence signals from 6–10 captured metaphase cells were combined to obtain an average ratio of tumor to normal DNA for each sample. Thresholds above 1.2 or below 0.8 defined gain or loss of genetic material, respectively. The results focused on specific chromosomal losses or gains in 18 primary and metastatic tumors, as well as on the degree of similarity (clonality relationship = CR) between individual primary and metastasis pairs. The aggregated findings revealed

that CGH abnormality profiles were similar for the primary tumors and their pulmonary metastases. The pulmonary metastases, however, contained more CGH abnormalities. They further observed that individual primary and metastasis pairs with high degrees of CR carried more 8p loss in the primary tumors and more 4q loss in the metastases compared to the low-CR group. The authors concluded that primary colon cancers with more genomic lesions, especially 8p and 4q losses, were more likely to metastasize.

Using similar methodologies and subject matter, **Tanami *et al.***² in this issue of *Laboratory Investigation* identified gains in DNA copy-number in chromosome 6p in liver metastases as compared to their colorectal primary. Upon further analysis using CGH-array techniques, CCND3 or Cyclin D3 was the only amplified gene identified after screening colorectal cell culture lines. Quantitative RT-PCR and immunohistochemical staining detected significant upregulation of CCND3 in samples of liver metastases. Expression of CCND3 was then found to correlate (negatively) with clinical outcomes of

patients with primary non-metastatic colorectal carcinoma.

In summary, using new and powerful techniques such as CGH, these authors were able to screen the entire genome and identify genetic sequences unique to the metastatic phenotype. This approach can serve as a broad platform to further investigate individual genes and proteins that are involved in this important and complex transformation.

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References

- 1 Jiang J-K, Chen Y-J, Lin C-H, *et al.* Genetic changes and clonality relationship between primary colorectal cancers and their pulmonary metastases—an analysis by comparative genomic hybridization. *Genes Chromosomes Cancers* 2005; 43:25–36.
- 2 Tanami HT, Tsuda H, Okabe S, *et al.* Involvement of cyclin D3 in liver metastasis of colorectal cancer, revealed by genome-wide copy-number analysis. *Lab Invest* 2005;85:1118–1129.