

**THE MODULATION OF IRON REGULATORY PROTEIN 1 (IRP1) BY  
NITRIC OXIDE (NO) IN A PAIR OF MOUSE LYMPHOMA CELL  
LINES L5178Y DISPLAYING DIFFERENT CONTROL OF IRON  
METABOLISM**

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Iron and nitric oxide are intimately associated in various biological processes. It is now well established that nitric oxide is one of the major pathophysiological stimuli that modulate the activity of iron regulatory protein 1 (IRP1), a key effector molecule involved in the regulation of intracellular iron metabolism. IRP1 is a cytoplasmic aconitase when it contains a [4Fe-4S] cluster, and an RNA-binding protein after complete removal of the metal center. By binding to specific mRNA sequences termed iron responsive elements (IREs), IRP1 modulates ferritin mRNA translation and transferrin receptor stability.

In this study, we used a pair of mouse lymphoma cell lines L5178Y which exhibit two different patterns of iron metabolism, i.e. the LY-R parental line with a high IRP1 and a high labile iron pool (LIP) level *versus* the LY-S mutant line with low IRP1 and LIP levels [Lipiński et al., *Blood* **95** (2000) 2960]. Cells were exposed to NO either via a 2-hour incubation with a NO donor, SpermineNONOate, or a 2-hour co-culture with mouse RAW 264.7 macrophages previously activated for NO synthesis. After the end of exposure to NO, the cells were analysed for both the aconitase and the RNA-binding activity of IRP1. Using these two different sources of NO, we observed a rapid, 2-3-fold increase in IRP1 RNA-binding activity in the two cell lines, as compared to the untreated controls. However, NO-induced activation of IRP1 observed in LY-R cells was twice as high as in LY-S cells. Simultaneously, we observed a loss of aconitase activity for IRP1, again, much more pronounced in LY-R than in LY-S cells.

Research is in progress to understand the biological mechanism(s) of the different ability of NO to activate IRP1 in the two L5781Y cell lines.