LETTER

Does casein kinase II phosphorylation of Maf1 trigger RNA polymerase III activation?

The recent study of Graczyk et al. (1) reported a role for casein kinase II (CK2) phosphorylation of Maf1 during the reactivation of RNA polymerase III (Pol III) transcription that occurs as cells transition from repressive to favorable growth conditions. This is a potentially important finding. However, the preceding conclusion remains in question because of two concerns that allow alternative explanations for the data.

Using the differential mobility of Maf1 in one-dimensional SDS gels, Graczyk et al. (1) reported changes in Maf1 phosphorylation upon conditional or pharmacological inhibition of CK2. However, prior work has established that these same changes in the mobility of Maf1 can be caused by mutation of as few as two phosphorylation sites (serines 177 and 178), sites targeted by protein kinase A and Sch9 that are not part of a CK2 recognition motif (2). The Graczyk et al. study did not determine the actual sites that were differentially phosphorylated. Rather the authors relied on a potentially confounding result to support their case, namely, that five serine-to-alanine substitutions at known or suspected CK2 sites altered Maf1 mobility. An alternative explanation is that structural changes in Maf1 caused by these five mutations altered the phosphorylation of serines 177 and 178 and/or the other five PKA/Sch9 sites (2, 3) independent of any involvement of CK2.

In addition, many experiments used a common design whereby cells grown in glycerol (repressing medium) were returned to glucose (activating medium) with or without simultaneous inhibition of CK2 activity. This experimental design was used to demonstrate changes in Maf1 phosphorylation, Maf1 association with tRNA genes, Maf1 interaction with Rpc160, and Pol III transcription. A consistent pattern of results was obtained, and the failure to restore normal Maf1 behavior or transcription when CK2 was inhibited was inferred to reflect the action of CK2 on Maf1. However, CK2 activity is essential for viability, and its inhibition has many effects on cellular function (4) that could lead to secondary or indirect consequences for Maf1 and Pol III transcription. Thus, the nutritional stress of growth in glycerol may be replaced by the cellular stress of greatly limited CK2 activity. Cellular stress due to the deficiency in CK2 could account for the failure to relieve Maf1-dependent repression of Pol III transcription.

We believe that until more research comes to light, the claim that CK2 is a direct, physiological regulator of Maf1 function during the activation of Pol III transcription is controversial.

***Ian M. Willis*[a,b,1], *Robyn D. Moir*[a], *and Jaehoon Lee*[a]**
Departments of [a]*Biochemistry and* [b]*Systems and Computational Biology, Albert Einstein College of Medicine, Bronx, NY 10461*

1. Graczyk D, et al. (2011) Casein kinase II-mediated phosphorylation of general repressor Maf1 triggers RNA polymerase III activation. *Proc Natl Acad Sci USA* 108:4926–4931.
2. Lee J, Moir RD, Willis IM (2009) Regulation of RNA polymerase III transcription involves SCH9-dependent and SCH9-independent branches of the target of rapamycin (TOR) pathway. *J Biol Chem* 284:12604–12608.
3. Huber A, et al. (2009) Characterization of the rapamycin-sensitive phosphoproteome reveals that Sch9 is a central coordinator of protein synthesis. *Genes Dev* 23:1929–1943.
4. Poole A, et al. (2005) A global view of CK2 function and regulation. *Mol Cell Biochem* 274:163–170.

Author contributions: I.M.W., R.D.M., and J.L. analyzed data; and I.M.W. wrote the paper.
The authors declare no conflict of interest.

[1]To whom correspondence should be addressed. E-mail: ian.willis@einstein.yu.edu.