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**THE REGULATION OF GLYCOGEN METABOLISM IN THE YEAST
*SACCHAROMYCES CEREVISIAE***

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ABSTRACT

Glycogen synthase (GS) catalyzes the biosynthesis of glycogen, and is controlled by reversible phosphorylation. In the yeast *Saccharomyces cerevisiae*, a type I protein phosphatase, Glc7p, dephosphorylates GS. A mutation in the *GLC7* gene (*glc7-1*) diminishes glycogen synthesis due to defective dephosphorylation of GS.

Therefore, I searched for second site suppressors of *glc7-1* to screen for new genes which might influence the phosphorylation of GS. A suppressor was isolated and identified as a mutation in *REG1*, a negative regulator of glucose repression. A *reg1* deletion also caused hyperaccumulation of glycogen. Epistasis analysis indicates that, as in the glucose repression pathway, *REG1* also acts upstream of the gene *SNF1* in the regulation of glycogen metabolism.

The *SNF1* gene encodes a protein kinase necessary for expression of glucose-repressible genes and for the synthesis of glycogen. The impaired glycogen synthesis in *snf1* cells is due to inactivation of GS. In another genetic screen, I found that mutation of the *PFK2* gene, which encodes the b subunit of 6-phosphofructo-1-kinase, restores glycogen accumulation in *snf1* cells. Deletion of *PFK2* from wild-type cells causes elevated level of glucose-6-P, hyperaccumulation of glycogen and activation of GS. Glucose-6-P is an allosteric activator of GS but additionally affects its phosphorylation. GS kinase activity was reduced in extracts from *pfk2* cells but was close to that of wild-type if the extract was gel-filtered to remove small molecules. Added glucose-6-P inhibited the GS kinase activity half-maximally at ~2 mM.

Another second site suppressor of the glycogen defect of *snf1* cells is *PHO85*, which encodes a cyclin-dependent protein kinase. Cells disrupted for *PHO85* showed a glycogen phenotype, including hyperaccumulation of glycogen, activation of GS and impaired GS kinase activity. Analysis of Ser to Ala mutations at the three potential Gsy2p phosphorylation sites in *pho85* cells implicated Ser654 and/or Thr667 in *PHO85* control of GS. Two Pho85 cyclins, Pcl8p and Pcl10p, were found to direct Pho85p to phosphorylate GS. Disruption of both *PCL10* and *PCL8* genes results in the same glycogen phenotype as in *pho85*, but does not show other phenotypes, suggesting that Pcl8/10p direct Pho85p specifically to GS. It is proposed that Pho85p-Pcl8/10p is a physiological GS kinase complex.

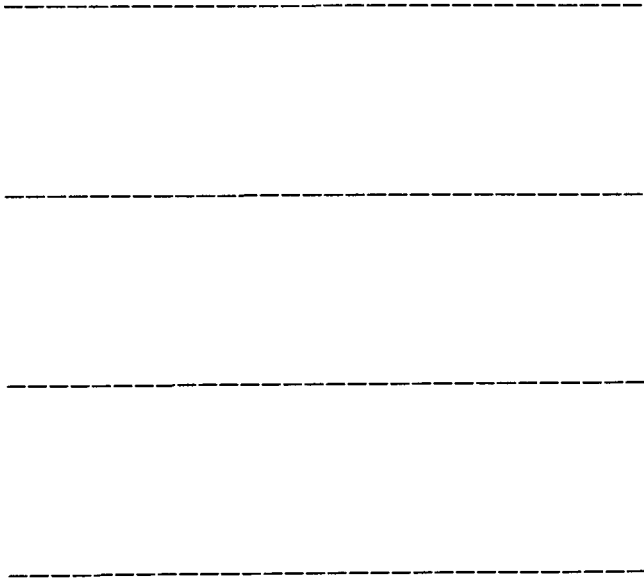


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