DBF4 is an essential gene that is expressed in a cell cycle-dependent manner. It encodes the regulatory subunit for the protein kinase Cdc7p; the activity of the Cdc7p-Dbf4p complex is required throughout S phase for the initiation of DNA synthesis at replication origins. The function of the Cdc7p-Dbf4p complex is analogous to that of cyclin-CDK complexes; here, Cdc7p is the catalytic subunit and is activated by association with Dbf4p during late G1. The Cdc7p-Dbf4p complex interacts physically with replication origins: Cdc7p interacts with Orc2p, and Dbf4p interacts with origin DNA. Dbf4p interacts with replication origins and with Cdc7p through different domains; both domains are essential for its function. Several lines of evidence suggest that members of the Mcm2-7 protein family are the most likely candidates for in vivo Cdc7p-Dbf4p substrates. Mcm2p and other Mcm2-7 proteins are in vitro substrates for Cdc7p-Dbf4p kinase activity, and the phosphorylation of Mcm2p in vivo depends upon Cdc7p-Dbf4p. Further, a mutation in MCM5/CDC46 can bypass the requirement for Cdc7p kinase activity in initiating DNA synthesis. Results of two-hybrid and GST-Mcm2 fusion affinity column chromatography showed that Cdc7p and Dbf4p interact physically with Mcm2p 6. A homolog of Dbf4p has been identified in S. pombe, and homologs of Cdc7p have been found in S. pombe, Xenopus, and human; evidence suggests that the homologs may perform the same function as the S. cerevisiae proteins.