SL304

The Regulation Mechanism of Chitin Synthetases in *Saccharomyces cerevisiae*

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The three chitin synthetases of *Saccharomyces cerevisiae*, Chs1, Chs2, and Chs3, participate in septum and cell wall formation of vegetative cells and in wall morphogenesis of conjugating cells and spores. Because of the differences in the nature and in the time of execution of their functions, the synthetases must be specifically and individually regulated. The nature of that regulation has been investigated by measuring changes in the levels of the three synthetases and of the messages of the three corresponding gnes, *CHS1*, *CHS2*, and *CAL1/CSD2/DIT101*(referred to below as *CAL1*), during the budding cycles. For Chs1 and Chs3, posttranslational regulation, probably by activation of latent forms, appears to be predominant. Since Chs2, like Chs1, is found in the cell in the zymogenic form, a posttranslational activation step appears to be necessary for this synthetase also.

The regulation mechanism was investigated to search the relationship of *CAL1*, *CAL2* and *CAL3* which is involved in Chs3 activity using different assay methods other than previous one. Treatment of Chs3-containing membranes with detergents drastically reduced the enzymatic activity. Activity could, however, be restored by subsequent incubation with trypsin or other proteases in the presence of UDPGlcNAc. Experiments with mutants in the three genes involved in Chs3 activity-*CAL1*, *CAL2*, *and CAL3*-showed that only *CAL1* and *CAL3* are required for the protease-elicited(zymogenic) activity. It is concluded that Chs3 is a zymogen and that the *CAL2* product functions as its activator.