Yeast catalase assay

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Abstract

Simple assay to see the effect of microcystin on yeast catalase activity. Important note is to handle solutions containing microcystin only with glassware (no plastic pipettes, microcystin sticks to plastic).

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Protocol

Step 1.
Make multiple liquid cultures of yeast cells and grow them overnight on shaking (230rpm) in 30 °C

Step 2.
Spin the cells down and discard supernatant, resuspend cells in 5 ml of 2x PBS and measure (and mark down) OD600. Be sure that there is more than 1,8*10^7 cells in the solution meaning OD has to be >1; more cells means a bigger, more easily measured response. When measuring OD do a dilution so that the value you get is between 0,1-0,8, and calculate the real OD of original sample.

Step 3.
Pipette 0,5 ml of cell/saline solution into each glass tube, add 0,5 ml of microcystin solution and mix properly -> leave into room temperature, 1 h - 4 h.

Step 4.
Add 0,5 ml of 30 % H2O2 and 0,2 ml of 1 % TritonX-100 solutions.

Step 5.
Wait 5 minutes and measure the foam layer. Foam layer height divided by width is used as a measure of catalase activity.