

Mikrobiologiya 2004 vol.73 N6, pages 734-740

Analysis of the Ca²⁺ response of mycelial fungi to external effects by the recombinant aequorin method

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Abstract

Using the mutant strain *Aspergillus awamori* 66A producing a recombinant Ca²⁺-dependent photosensitive protein aequorin, the dynamics of Ca²⁺ was studied for the first time in the cytosol of the micromycetes exposed to stressful factors, such as an increase in extracellular Ca²⁺ to 50 mM, hypoosmotic shock, and mechanical shock. Cell response to stress proved to involve an increase in the Ca²⁺ concentration in the cytosol, which was determined from the amplitude of aequorin luminescence and the time of the amplitude enhancement and relaxation. The level of Ca²⁺ response depended on the physiological stimulus. Inhibitory analysis with various agents that block Ca²⁺ channels and with agonists that specifically enhance the activity of the channels suggested that (1) the level of Ca²⁺ in the cytosol of micromycetes increases in response to stress because of the ion influx from both the growth medium and intracellular reservoirs and (2) the potential-dependent transport systems play the major role in the Ca²⁺ influx into the cytosol of the micromycete cells.

Keywords

Aspergillus awamori, Ca²⁺ dynamics, Mycelial fungi, Recombinant aequorin, Stress