

Characterization of tolaasin inhibitory factors for the suppression of brown blotch

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The brown blotch disease of cultivated oyster mushroom is evoked by tolaasin, a 1.9 kDa peptide. Tolaasin is produced by *Pseudomonas tolaasii* and forms pores in the cellular membranes of cultivated mushroom. Previously, we showed that the tolaasin-induced pore formation required the multimerization of tolaasin molecules and the multimerization of tolaasin was blocked by the treatment of various chemical compounds. These chemicals were named tolaasin-inhibitory factor (TIF) to prevent the brown blotch disease. Among different food additives, fatty acid derivatives blocked effectively the tolaasin-induced hemolysis. These compounds were effective at concentrations of 10-100 μM and sometimes even at 1 μM . TIF successfully inhibits the ion channel activities of tolaasin channel in artificial lipid bilayers. TIF activities were not dependent on temperature and pH, although haemolytic activity of tolaasin increased at higher temperature. Chemical properties of TIF were investigated to develop a commercial product for farmers. Since TIFs are unsaturated carbon compounds, they were sensitive to the air exposure and light irradiation. In nitrogen-filled air tight containers, TIFs were stable and less than 10% activity was decreased during a storage period of three months. However, more than 90% of TIF activity was suppressed by two weeks in the presence of oxygen. Room light also decreased the TIF activity by two times. Temperature did not show significant effects on the stability of TIFs, since storage at 5, 25, and 45°C did not show any difference. Therefore, in order to make the storage period of TIF longer, containers should be designed to oxygen-tight and light-free. Currently, the degradation products of various TIF are being identified by mass spectroscopy to analyze solution the structure of tolaasin and binding sites of TIF.

