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Native and heterologous protein oxidation and subsequent degradation in a recombinant filamentous fungus *Aspergillus niger* B1-D

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Filamentous fungi have attracted extensive research interest due to their abilities to secrete large amounts of high-valued recombinant proteins. Despite the fact that proteases have been recognized as one of the main problems in protein production, little is known about the regulation of them. In the present study, we hypothesized that reactive oxygen species (ROS), unavoidable by-products in all aerobic cultures, may cause protein oxidation and induce proteolysis in filamentous fungi. To test this hypothesis, we used a variety of oxidative stressors, oxygenation, menadione, hydrogen peroxide, hydrogen peroxide with copper ion, to study the induction of intracellular proteolytic activities in *A. niger* B1-D, a recombinant filamentous fungus secreting hen egg white lysozyme. Protein carbonyl content was monitored as a bio-marker for protein oxidation. Our results show that oxygenation and metal-catalyzed oxidation significantly induce carbonyl groups to bovine serum albumin (BSA). We also found that proteolytic activities and carbonyl content increase on the addition of these stressors to the whole broth in *A. niger* B1-D, and the responses are dose-dependent. In conclusion, ROS overproduction correlates with protein oxidation and proteolysis in *A. niger* B1-D. It is advisable to decrease ROS production in the bioprocess of filamentous fungi in order to achieve a higher productivity of heterologous proteins.