

Gene-Protecting Properties of Melanin Pigments in Medicinal Mushrooms

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Melanin pigments synthesized by mycelial fungi, especially Basidiomycetes, are not regarded as an attractive object for study although they may serve as readily available and economically acceptable feedstock for the pharmaceutical industry. Our investigation was aimed at recovery of melanins from cultural liquid and mycelia of fungi *Inonotus obliquus* (Pers.) Pil. and *Phellinus robustus* P. Karst. to trace their gene-protective potential in the cause of metabolic activation of *o*-dianisidine (*o*-DA, 3,3'-dimethoxybenzidine) via a peroxidase oxidation pathway. Metabolic activation of DA and other aminobiphenyls generates electrophilic products able to interact with nucleophilic groups in biopolymers, in particular DNA, yielding intermolecular DNA-DNA links. A major product of *o*-DA peroxidase oxidation is diimine, responsible for DNA crosslinking.

Introduction into the reaction mixture of melanins derived from both mushroom species at concentrations > 20 mg/ml inhibited the process. It is noteworthy that the concentration of pigment produced from *Ph. robustus*, reducing the *o*-DA oxidation rate twofold, equaled 135 mg/ml compared to 85 mg/ml for *I. obliquus*, indicating the higher peroxidase inhibition potential of the latter.

To evaluate the gene-protective effect of the pigments on λ phag DNA lesions induced by products of *o*-DA peroxidase oxidation, DNA damage was correlated with the initial *o*-DA level. Concentrations exceeding 0.03 mM resulted in 100% DNA damage.

Pigment content in *Ph. robustus* sufficient to decrease DNA crosslinking twofold constituted 3.1 mg/ml, whereas the figure for *I. obliquus* reached 6.0 mg/ml. Melanin concentrations of 10 mg/ml and 20 mg/ml respectively, completely suppressed the process. Control melanin isolated from the mushroom *Aspergillus carbonarius* (Bain.) Thom (for comparison) showed a 56% DNA-protective effect only at 20 mg/ml reaction mixture, while 100% blocking of lesions occurred at 60 mg/ml concentrations.

Antimutagenic activity of mushroom melanins was tested on male Af mice. Experiments demonstrated that intraperitoneal injection of *A. carbonarius* pigment at a dose of 50 mg/kg body weight decreased radioinduced spermatocyte translocations by 1.5 times whereas in the case of *I. obliquus* the melanin effect was twofold.

These findings indicate the high gene-protective potential of mushroom melanins both *in vivo* and *in vitro*.