OXYGEN CONCENTRATION ALTERS GENE EXPRESSION AND MANNITOL SYNTHESIS OF *Beauveria bassiana*

Paul Misael Garza-López[1], Aida Hamdan-Partida[2], Octavio Loera[1]. [1]Depto. de Biotecnología, UAM-Iztapalapa, C. P. 09340, México, D. F. [2]Depto. de Sistemas Biológicos UAM-Xochimilco, C. P. 04960, México, D. F. Email: paulmisael@gmail.com

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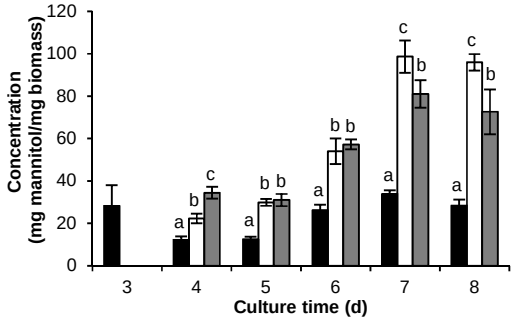
Introduction. The entomopathogenic fungus *Beauveria bassiana* is a microorganism used as a biological control agent. As an aerobic, this fungus produces reactive oxygen species (ROS), which can be eliminated by action of compatible solutes such as mannitol [1]. The aim of this study was to determine the response in mannitol synthesis and in the *mpd* gene expression, encoding the enzyme mannitol-1-phosphate dehydrogenase, in *B. bassiana* under pulses of different O_2 concentrations, linked to an increased conidiation [2].

Methods. The strain of *B. bassiana* Bb 885.2 was subjected to pulses of three concentrations of O_2: 16%, normal atmosphere (21%, NA) and 26%, in solid cultures using rice as a substrate. These pulses started after 3 days of culture. Mannitol concentration was determined by HPLC and differential *mpd* gene expression was quantified by real time PCR (qPCR) [3, 4].



Fig, 1. Mannitol concentrarion of strain Bb 882.5 in cultures with different atmospheres. (■NA; □16% O_2;▩ 26% O_2). Letters distinguish significantly different values on each day (Tukey: $P < 0.05$).

Results. Mannitol concentration increased by at least 86% after modification of O_2 concentration, compared to NA (Fig. 1). Maximum concentration of the polyol was reached on day 7 in all treatments. At day 8, the highest concentration was obtained with 16% O_2 (100 µg mannitol/mg biomass) which was three times higher than the value obtained with 21% O_2 (30 µg mannitol/mg biomass) ($P < 0.05$). This is consistent with previous studies that reported an increase in mannitol concentration when *B. bassiana* was subjected to different types of stress [3]. Moreover, *mpd* gene expression was significantly higher at 16% and 26% treatments than in 21% O_2 (Fig. 2) ($P < 0.05$). For this quantification, days analyzed were: the first day of atmospheric modification (day 3), 24 h later (day 4) and the time at which mannitol concentration was considerably higher (day 6) (Fig. 1). Furthermore, a relationship between *mpd* relative gene expression and mannitol synthesis was observed, since both parameters were increased due to modified O_2 concentration (Fig. 1 and 2). In a previous study, the *mpd* gene deletion in *B. bassiana* caused significant decrease in the mannitol synthesis in normal culture conditions [4].

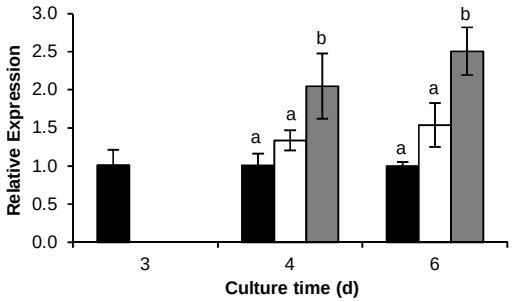


Fig. 2. Normalized relative expression of *mpd* gene of strain Bb 882.5 in cultures with different atmospheres. (■NA; □16% O_2; ▩26% O_2). Letters distinguish significantly different values on each day (Tukey: $P < 0.05$).

Conclusions. Mannitol synthesis increased due to modification of oxygen concentration in solid cultures which is directly related to the expression of *mpd* gene.

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