



CONIDIATION RESPONSE by *Metarhizium anisopliae* UNDER OXIDATIVE STRESS: SOLID vs. SUPERFICIAL CULTURES.

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Introduction. Most procedures for production of biocontrol agents such as *Metarhizium anisopliae* require additives like extract yeast, even chitosan [1], which increases the production cost. As an enhancer of conidial yields, oxygen pulses induce the conidiation on *M. anisopliae* [2]. The aim of this work was to select a substrate and then evaluate the effect of the oxygen pulses (26% O₂) on the conidiation of *M. anisopliae*, either under solid state or superficial culture.

Methods. *Metarhizium anisopliae* CP-OAX was grown on three media: oat, rice and oat-rice grain mixture (50%-50%). Conidia were evaluated after 4 and 7 days, measuring the conidial yields per gram of initial dry substrate (con/gds), and the best substrate was selected for further analysis. Then, *M. anisopliae* was grown on two systems: Solid State Culture (SSC) using the selected media in form of grains [3]. For the superficial culture, a fine powder of these grains was used as substrate in agar media [2]. Pulses of 26% oxygen were applied following the methodology described previously [2, 3]. Conidia production was evaluated daily and expressed as con/gds and con/cm² in SSC and superficial culture, respectively. Experiments were performed by six replicates.

Results. Solid State Cultures were performed in order to select the medium that produced the highest conidiation. The medium containing oat and rice (50%-50%) showed the best conidial yield reaching 1.91x10⁹ con/gds (Fig 1).

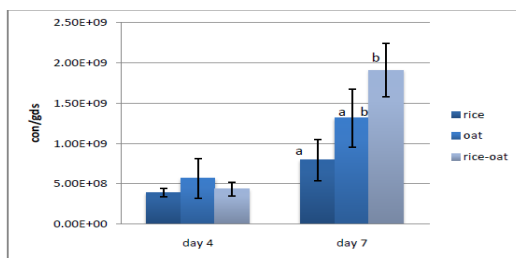


Fig. 1. Conidiation by *M. anisopliae* on SSC for medium selection. Letters distinguish significantly different values (Tukey P<0.05).

In the superficial cultures, the maximal conidiation was obtained with 26% O₂ pulses,

at 132 h (2.6x10⁷ con/cm²). This is in accordance with previous reports [2], who reached a level of 1.28x10⁸ con/cm² after 8 d. In the SSC, the O₂ pulses (26%) had a negative effect on conidiation compared to the control (21% O₂) (Fig. 2). This was similar to results previously observed in SSC with *B. bassiana* [3]. The differential effect of an oxidant condition as dependence of the culture system has not been described. This could be a consequence of the oxygen consumption rate in each system, which in turn could be related to an oxidative stress [4].

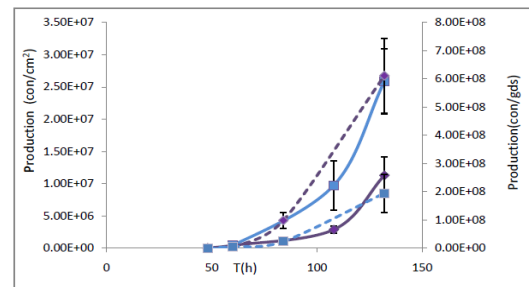


Fig. 2. Conidiation of *M. anisopliae* in two different systems. Solid State Cultures (dotted lines) and Superficial Cultures (solid lines). Normal atmosphere (rhombs) and 26% O₂ pulses (squares).

Conclusions. The effect of the oxygen pulses (26%) depended on the culture system, which should be considered as a designing factor for the production of conidia by *M. anisopliae*.

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