

Metarhizium anisopliae protease production in different culture media

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Control of pests and nematodes requires the massive use of chemical pesticides. In addition, the improper use of agrochemicals can harm the environment and human health. In view of this, alternative methods of disease and pest control are being sought. In this context, a current option is the use of entomopathogenic fungi, which carry out the infection process through two main approaches: mechanical, due to the pressure of the hyphae that breaks the membrane or slightly sclerotic areas; and biochemical, resulting from the production of extracellular hydrolytic enzymes such as proteases (EC 3.4). These enzymes are important for the degradation of the host cuticle. *Metarhizium* spp. stands out as one of the most used fungi genera, and is considered an alternative to the use of chemical pesticides. Among the challenges for its use, the use of an appropriate culture medium can be cited. For this reason, the present study aimed to evaluate the commercial isolates *M. anisopliae* IBCB 425 and *M. anisopliae* ESALQ E9 in different solid and liquid culture media for the production of proteases. *M. anisopliae* isolates were acquired as commercial products. *M. anisopliae* IBCB 425 was provided by Nitro Agro as Metarriz® and *M. anisopliae* ESALQ E9 was acquired from Koppert as Metarril®. These isolates were grown in Potato Dextrose Agar (BDA) 2.0 % (w/v), at 25 ± 1 °C, in the dark, for 10 days. The concentration of 10⁷ conidia/mL was used. Then, the conidial suspension was used for inoculation in liquid and solid media for the induction of protease production, for statistical analysis, we used the Tukey test at 1% probability. The isolated IBCB 425 (52 U/mg) showed significant difference (p < 0.01) from the rice-whey medium compared to the other media evaluated. The isolated *M. anisopliae* ESALQ E9 produced greater proteolytic activity using the media with soluble starch and yeast extract (16 U/mg) and rice-whey (16 U/mg). It was concluded that the isolates of the entomopathogenic fungi *M. anisopliae* IBCB 425 and ESALQ E9 used in the present study successfully produced protease, reaching high levels of activity. In addition, it was demonstrated that after evaluating various culture media, for all the isolates tested, the medium that induced the highest protease production was the rice-whey medium.

Keywords: Protease; Biological control; Culture medium; *Metarhizium anisopliae*.

Produção de proteases por *Metarhizium anisopliae* em diferentes meios de cultura

Metarhizium anisopliae produz enzimas hidrolíticas, principalmente, proteases (EC 3.4), que são fundamentais no processo de infecção dos organismos alvo. Assim, o presente estudo teve como objetivo avaliar os isolados comerciais *M. anisopliae* IBCB 425 e *M. anisopliae* ESALQ E9 em diferentes meios de cultura para a produção de protease. Foram inoculados 10⁷ conídios/mL nos diferentes meios de cultura. O isolado IBCB 425 (52 U/mg) produziu maior atividade de protease com o meio de soro de arroz de leite, em comparação ao *M. anisopliae* ESALQ E9.

Palavras-chave: Protease; controle biológico; meio de cultura; *Metarhizium anisopliae*.

Acknowledge: This work was carried out with the support of Minas Gerais State Research Foundation (FAPEMIG), National Council for Scientific and Technological Development (CNPq), Department of Chemistry of the Federal University of Lavras, Nitro Agro and Koppert.