

## STANDARDIZATION OF A CULTURE MEDIUM FOR RECOVERING *Pythium insidiosum* FROM ENVIRONMENT

VIRIATO, V.<sup>1</sup>; VIRIATO, A.<sup>2</sup>; BAGAGLI, E.<sup>1</sup>; BOSCO, S.M.G.<sup>1</sup>

<sup>1</sup>- Depto. de Microbiologia e Imunologia, Instituto de Biociências de Botucatu, UNESP

<sup>2</sup>- Instituto de Infectologia Emílio Ribas, São Paulo

*Pythium insidiosum* is the causative agent of pythiosis, a disease that affect animals including the human specie, mostly in tropical and subtropical regions. The pathogen is a fungus like organism, because it does not belong to Kingdom Fungi. The life cycle involves parasitism of plants in aquatic environment, where the production of infective form (biflagellate zoospores) occurs. There are few environmental studies on this pathogen, which was isolated from water samples in Thailand, a place that concentrates the highest incidence of the disease in humans. In Brazil, there are reports of isolation of *Pythium insidiosum* only in clinical samples of equines, dogs, ruminants and one human, however the isolation of the pathogen from water environments has not been reported in literature yet. There are some reports, mainly from São Paulo State, of the isolation of *Pythium echinulatum*, *Pythium graminicolum*, *Pythium irregulare*, *Pythium rostratum*, *Pythium spinosum*, *Pythium torulosum*, *Pythium ultimum*, *Pythium undulatum* and *Pythium vexans*. Soil and water environments have a great diversity of microorganisms, some of them are good competitors and can difficult the isolation of *P. insidiosum*. So, we aimed to standardize a culture medium that selects the growth of *P. insidiosum* by reducing and/or eliminating other fungal contaminants, such as *Aspergillus* spp. It was tested one culture of *P. insidiosum* (isolated from equine) and three isolates of *Aspergillus* spp. from environment: *A. fumigatus*, *A. flavus* and *A. niger*. These fungi were cultivated in Sabourad agar (SAB), considered as control group for the growth of fungi, and in the MIPi (Medium for Isolation of *Pythium insidiosum*), which consisted by glucose, peptone, antibiotics (chloramphenicol and gentamicin) and antifungals (amphotericin and derosal). Standardized fragments of 5 mm of diameter of the fungi were cultivated in both media and the growth was evaluated daily for seven days. It was observed that in SAB medium all fungi tested reached the edges of the Petri dishes, while in MIPi, only *Pythium insidiosum* grew. The ecological niche of *P. insidiosum* is still poor understood, however we believe that it is expanding even more, as a consequence of environmental changes. The survey of the pathogen in environment is important to map the risk areas for the disease and may allow for preventive measures to avoid new infections. We suggest the use of MIPi medium as selective for environmental studies in Brazil.

Palavras-chave: *Pythium insidiosum*, environment, selective medium, pythiosis.

Apoio: FAPESP