CUANTIFICATION OF PROTEIN AND PROTEASE BY *FOMES* sp EUM1 UNDER THEMAL STRESS

Martínez-Valdez Francisco[1], Ordaz Armando[1], Fernandez-Perrino Francisco[1], Díaz –Godínez Gerardo[2], Loera Octavio[1], [1]Universidad Autónoma Metropolitana-Iztapalapa, Departamento de Biotecnología, México, D.F., CP 09340; fco.jav.mv@gmail.com

Key words: Fomes, Protease, Stress Temperature

Introduction.

The genus *Fomes* belongs to the subdivision Basidiomycota, family Polyporaceae. Optimal growth temperature for this strain is 30°C although it showed thermotolerance traits (1). The stress by temperature induces the production of proteases, which catalyze the rapid degradation of secreted proteins. The heat shock response may be the synthesis chaperones and proteases, aiding in the repair and recovery of proteins from the cell (2). The objective of this work was to determine the production of proteases on superficial cultures under thermal stress by *Fomes* sp. EUM1.

Methods.

Fomes sp EUM1 was grown using stover corn as substrate previously sterilized (25 min at 120 °C). Petri dishes (90 mm x 15 mm) with 30 ml medium (stover corn 40 g/L and Agar 15 g/L) were inoculated with a suspension of mycelium in the center of the dishes with a volume of 200 µL. The inoculum was distributed on the agar surface in order to obtain a lawn growth (**Fig. 1**). The dishes were incubated for 12 days at 35°C and one portion of these dishes at day 6 were changed to a temperature of 45°C (3). Soluble protein was determined by the method of Bradford, reported as milligrams of protein per gram of initial dry substrate (mg/gids). The protease activity was measured using casein degree Hammarsten as a substrate (4).

Results.

Figure 2 Show the pH and soluble protein. *Fomes fomentarius* under oxidative stress changed enzymatic profiles due to the generation of reactive oxygen species, similar to those produced under thermal stress (4). **Figure 3** shows the activity of the proteases at both temperatures (35 and 45°C). Loss of activity at 45°C could probably due to denaturation of the proteases (5).

Fig.1 Superficial growth of *Fomes* sp. EUM1

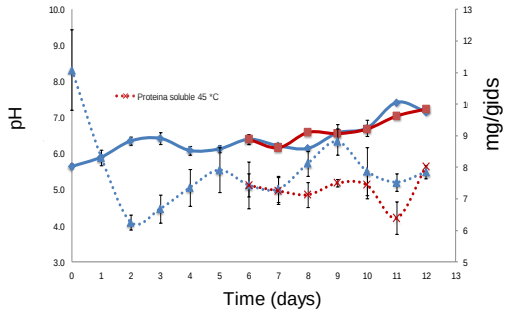

Fig.2 Measurement of pH (solid lines) at 35°C (blue) and 45°C (red) and quantification of soluble protein (dotted lines) at 35°C (blue) and 45° C (red).

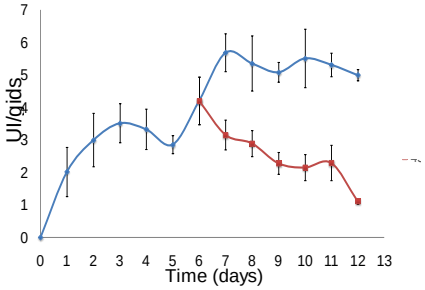

Fig.3 Proteases activity at 35°C (blue) and 45°C (red).

Conclusions.

The pH and soluble protein were less affected by thermal stress, although the strongest effect was observed with the proteolytic activities which could explain sustainable production of laccases enzymes at 45°C (3).

Acknowledgements. Authors thank to CONACyT for the scholarship (23705) and UAM-Iztapalalapa

References.

1. Ordaz *et al.* 2012. Hyphal morphology modification in thermal adaptation by the white-rot fungus *Fomes* sp. EUM1. *J Basic Microbiol* , 51, 1–8
2. Johri *el at*, 1985, Plant Sci. *Enzymes from thermophilic fungi: Proteases and lipases*. *Indian Acad. Sei*, India, Vol. 94, N°2 & 3
3. Martínez *et al.* 2011. *Thermal stress effects on the production of laccase by Fomes sp. EUM1*. MSc Theses in Biotechnology UAM-I.
4. Lee *et al* 1997. Alkaline serine protease is an exotoxin of Vibrio alginolyticus in kuruma prawn, Penaeus japonicus. *Curr Microbiol* 34:110–117.
5. Jaszek, *et al.* 2006, Enhanced extracellular laccase activity as a part of the response system of white rot fungi: *Trametes versicolor* and *Abortiporus biennis* to paraquat-caused oxidative stress conditions. *Pestic. Biochem. Phys.*, 85, 147-154