



## BIOCHEMICAL CHARACTERIZATION OF A THERMOPHILIC XYLANASE (XYL86) FROM THE THERMOPHILIC FUNGUS *CORYNASCUS SEPEDONIUM*

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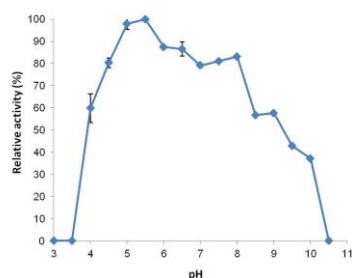
**Key words:** Xylanases, xylan degradation, *Corynascus sepedonium*

**Introduction.** Xylanases (EC 3.2.1.8) are the key enzymes for xylan degradation and they can be effectively used with cellulases to hydrolyze the lignocellulosic biomass, in order to produce bioethanol. These enzymes are produced by a variety of microorganisms including filamentous fungi (1). The thermophilic fungus *C. sepedonium* Co3Bag, isolated from sugar cane bagasse compost, produce lignocellulolytic activities (2). We have previously reported the purification of two thermophilic enzymes from *C. sepedonium* Co3Bag1: a laccase (3), and a xylanase (4). However, the catalytic properties of the xylanase, produced by this fungus, have not been studied so far.

The aim of this work was to carry out the biochemical characterization of a xylanase of 86 kDa (Xyl86) produced by *C. sepedonium* Co3Bag1.

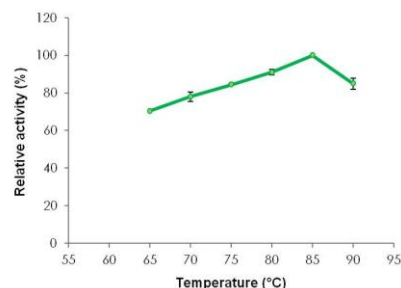
**Methods.** Xyl86 was purified from the culture supernatant of *C. sepedonium* Co3Bag1 as described (4). The optimal pH and temperature, thermal stability, the effect of several metal ions (1 and 5 mM), and the kinetic parameters  $K_m$  and  $V_{max}$  were determined using beechwood xylan as substrate, as described (5).

**Results.** Xyl86 showed an optimal activity at pH 5.5 and maintained over 80% of its maximum activity in a wide range of pH (4.5-9) (Fig. 1).



**Figure 1.** Effect of pH on Xyl86 activity from *C. sepedonium* Co3Bag1

Xyl86 displayed the highest activity at 85°C and pH 5.5, the optimal pH condition (Fig. 2).



**Figure 2.** Effect of temperature on the Xyl86 activity from *C. sepedonium* Co3Bag1

Studies on the effect of EDTA and different metal ions on the Xyl86 activity showed  $\text{Cu}^{+2}$ ,  $\text{Mg}^{+2}$ ,  $\text{Ni}^{+2}$  and EDTA reduced the enzymatic activity to different extents, while  $\text{Ca}^{+2}$  had no effect; however, the enzyme was completely inhibited by  $\text{Hg}^{+2}$ . Results obtained for the thermostability of Xyl86 and determination of  $K_m$  and  $V_{max}$ , are shown on Table 1.

**Table 1.** Thermostability and kinetic parameters of Xyl86 from *C. sepedonium* Co3Bag1

<b>Thermostability</b>	85 °C	$t_{1/2}$ 36 min
	75 °C	$t_{1/2}$ 230 min
<b>Kinetic parameters</b>	$K_m$	0.415 $\text{mg}\cdot\text{ml}^{-1}$
	$V_{max}$	0.896 $\mu\text{mol}\cdot\text{min}^{-1}\cdot\text{ml}^{-1}$

**Conclusions.** Xyl86 is an acid and thermophilic xylanase active in a wide range of pH and temperature, and is stable at temperatures higher than 70°C. Hence, Xyl86 might represent a suitable candidate for the enzymatic pretreatment of lignocellulosic biomass.

**Acknowledgements.** Research was funded by CINVESTAV, México. YGGH received a scholarship from CONACYT, México.

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