



PRODUCTION AND QUANTIFICATION OF CELLULASE FROM MICROORGANISMS ISOLATED FROM BOVINE RUMEN, USED TO DEGRADE NOPAL (*Opuntia Sp.*)

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Introduction. Enzymes are a special class of proteins which accelerate the rate of chemical reactions that occur in a cell. For this reason they are known as "biological catalysts". The mode of action is specific and each type of enzyme which acts on a particular type of reaction and on a specific substrate. Because the use of some enzymes of vegetable and animal origin has been decreasing, it has been chosen for the enzymes of microbial origin within which the cellulase was found, which is rapidly hydrolyzed in the nature, particularly the anaerobic organisms in the rumen and intestine, of ruminants (Carrera, 2003)

Methods. The strain VML-2 (ruminal microorganism) was isolated in a solid culture media "agar Scheadler", and then a liquid medium was prepared with the following specific components: NaCl, NaNO₃, KI, KH₂PO₄, MgSO₄, CMC (carboximetilcelulose) as a carbon source, and 7 ml of pure culture, were incubated at 40°C for 96 hours under anaerobic conditions. Medium was centrifuged at 5000 rpm for 10 minutes to determine protein and extracellular protein by the Biuret method. Then a cactus (*Opuntia sp.*) enzyme kinetics were following using the enzyme extract at times of 0=0min, 1=5min, 2=15min, 3=60min, 4=90min, 5=120min, 6=24h, 7=48h, 8=72h, and 9=96h. Reducing sugars was determined using the technique of Somogyi-Nelson.

Results. The amount of cellular and extracellular protein at 96 hours of fermentation was 95 mg/ml. Figure 1 shows the growth curve using CMC with an exponential phase of 12 to 72 hours with $\mu = 0.9963$ OD / h, then there is an immediate drop in the graph where microbial growth slows due to depletion of nutrients or the accumulation of toxic waste. The figure 2 shows the enzymatic activity of the cellulase

over *Opuntia sp.* It's observed that in the first 60 minutes of reaction time has the maximum activity with a value of 1430 U. Valdes (2010) reported enzymatic values of 858 U at 24 h of fermentation using *O. pheacanta*.

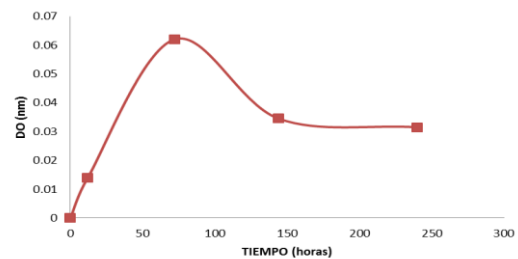


Fig 1. Growth curve in carboxymethyl cellulase incubated at 40 ° C.

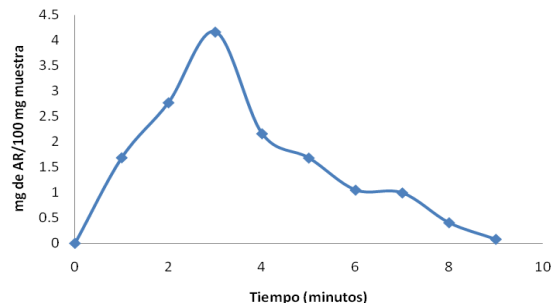


Fig 2. Enzymatic degradation of cellulose in *Opuntia sp.*

Conclusions. Use of conventional an economic substrates represent an alternative to produce cellulases using microorganisms of rumen.

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