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Short Communication

Influence of water activity on growth and activity of *Aspergillus niger* for glycoamylase production in solid-state fermentation

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Growth of *Aspergillus niger* and glucoamylase production correlated well with the water activity of the substrate (wheat bran plus corn flour) in a solid-state fermentation. Both were maximal at an initial water activity of 0.936. Glycoamylase reached 550 units/g dry substrate after 96 h.

Key words: Aspergillus niger, glucoamylase, solid-state fermentation.

Solid-state fermentation (SSF) has attracted interest because of its advantages and potential for the industrial production of useful microbial products (Pandey 1992). The water activity (aW) of the substrate is important in SSF because at relatively low moisture contents growth and metabolism of the microorganism can be limited. Control of aW could be used to modify metabolite production or secretion of a product (Gervais 1990). Our studies on SSF using *Aspergillus niger* NCIM 1245 have shown it has a good potential for producing high titres of glucoamylase. In this paper, we report how the aW of the substrate affects the growth of and enzyme synthesis by this strain.

Materials and Methods

Commercially available wheat bran of mixed particle size and corn flour were mixed and supplemented with a mineral solution (Pandey 1990). Glycerol was used to establish four experimental aW values: 0.936, 0.902, 0.819 and 0.742. Fermentation was carried out with 50 g wet substrate in a 500 ml wide-mouth conical flask and inoculated with spore suspension of *Aspergillus niger* NCIM 1245 as described earlier (Pandey 1990). Flasks were incubated at 30 ± 1°C for 120 h. Whole flasks were taken for each sample.

Glucoamylase was extracted from the fermented matter using distilled water and assayed using starch as substrate (Pandey 1990). Reducing sugars were determined by the dinitrosalicylate method. Glycoamylase units (IU) are expressed as μmol reducing sugars released per min by the total amount of enzyme extracted from 1 g dry substrate (g ds). Mycelial growth was determined by estimating glucosamine in the fermenting substrate (Sakurai *et al.* 1977). The substrate aW was measured using a water activity meter.

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Results

Figure 1 shows the growth profile of *A. niger* on substrates with different initial aW. Lower aW values adversely affected growth. With initial aW values of 0.936 and 0.902, mycelial growth continued to increase until 72 h (giving 16.7 and 14.3 mg glucosamine/g ds, respectively) and then declined. With initial aW values of 0.819 and 0.742, growth was three to five times less than on the substrate with an aW of 0.936. The aW values in all cases increased slightly as the fermentation proceeded (Figure 1). Values of aW over 0.97 are usually not inducive for good growth and activity of *A. niger* NCIM 1245 (data not shown), and the cessation of growth after 72 h on substrates with aW of 0.936 and 0.902 could be due to this. Decreased aW of the substrate can lead to lower mass transfers and water availability for the microorganism and may thus be responsible for an incomplete conversion of the substrate to biomass.

Figure 2 shows the activities of glucoamylase in the fermented substrate during the course of fermentation. At high

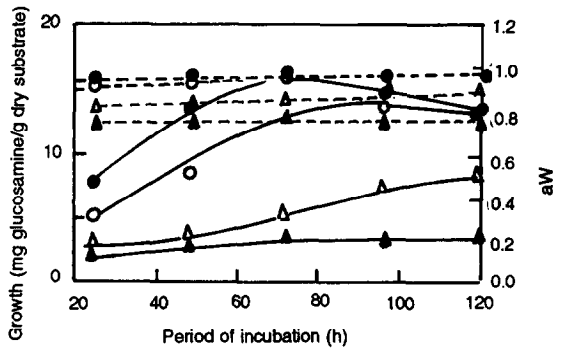

Figure 1. Growth of *A. niger* (——), measured as mg glucosamine/g dry substrate, at different aW values, and the change in aW of the substrate (- - - -) during solid-state fermentation. Initial aW was 0.936 (●), 0.902 (○), 0.819 (△) or 0.742 (▲).

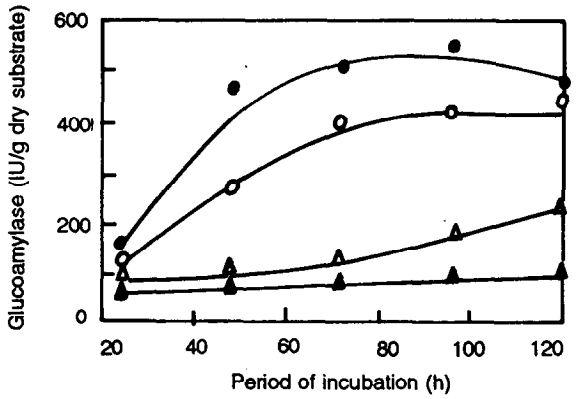

Figure 2. Glucoamylase activities during solid-state fermentation with initial aW values of 0.936 (●), 0.902 (○), 0.819 (△) or 0.742 (▲).

initial aW values the enzyme activities were relatively high (to maxima of 550 and 420 IU/g ds after 96 h at aW of 0.936 and 0.902, respectively). At aW of 0.819 and 0.742 the yields continued to increase until the end of fermentation, being maximum after 120 h (225 and 100 IU/g ds, respectively). These results correlate with the growth pattern given in Figure 1.

In a solid-state fermentation process, the aW of the substrate influences the enzyme activity and protein stability. Water acts as a vehicle for substrate transport and as a reactant so it may be expected that aW affects enzymatic transformation during fermentation. Our results are in accord with these views.

Acknowledgements

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