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Short Communication

Aldolase activity in *gal* mutants of *Aspergillus nidulans*

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Abstract

Aldolase assayed in some *gal* mutants of *A. nidulans*, when subjected to galactose toxicity under defined growth conditions, indicated that glycolysis is impaired.

Key words : Aldolase activity, galactose toxicity, glycolysis, galactosemia.

1. Introduction

Galactosemia is an inherited disorder of galactose utilisation, wherein one or more enzymes, involved in its metabolism, is deficient[1,2]. A similar situation is reported in mutants of microorganisms which are unable to utilise galactose as the sole source of carbon[3–6]. In this communication, we report the changes in the levels of aldolase activity in some *gal* mutants of *A. nidulans*. This study was undertaken to evaluate the toxic effects of this sugar and its metabolites upon the activity of this key enzyme in glycolysis in the " galactosemic state ".

2. Materials and methods

Six *gal* mutants and the wild strain of *A. nidulans* were used in the present investigation. The conditions of growth tested and the nature of the mutants are described in the legend to Fig. 1.

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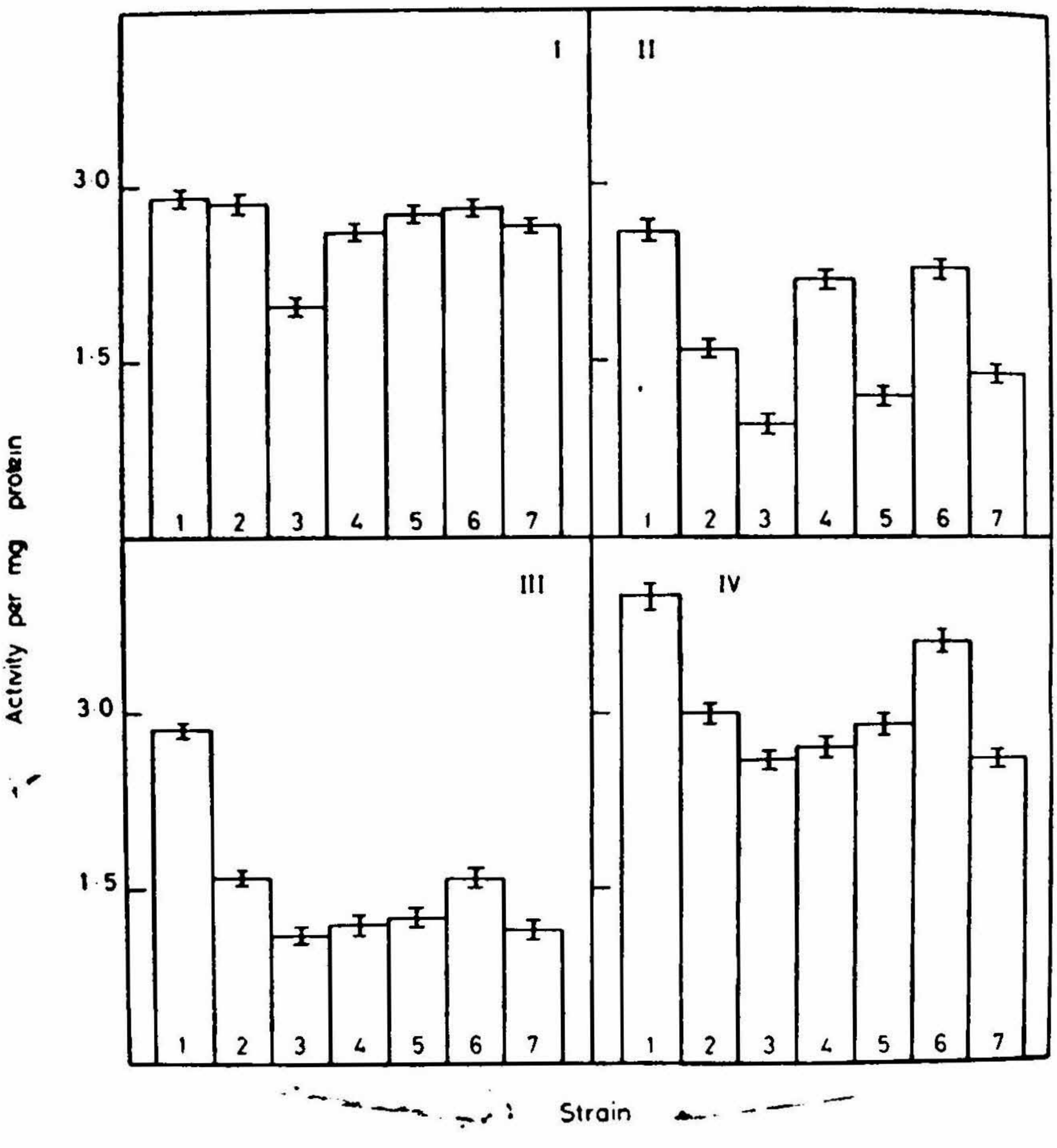

FIG. 1. Aldolase activity in the mycelia of the wild and mutant strains of *A. nidulans* grown in the media described, for 3 days at 37° C. Activity is expressed as mc moles of glyceraldehyde formed per mg protein.

I—Medium containing galactose and glucose in the ratio of 1 : 1.
II—Medium containing galactose and glucose in the ratio of 3 : 1.
III—Medium containing only galactose.
IV—Medium containing only glucose.

Key : 1—wild ; 2—gal_1 (constitutive) ; 3—gal_9 (kinaseless) ; 4—gal_3 (slow-growing) ; 5—gal_4 (slow growing) ; 6—gal_{10} (partial) ; 7—gal_5 (transferaseless).

Media were prepared according to the method described by Pontecorvo *et al*[6]. Fifty ml of the media were dispensed in 250 ml Erlenmeyer flasks, inoculated with the spore suspension of the strains (containing 10^8 spores/ml) and incubated for 3 days at 37° C. The mycelia were then harvested and extracted with buffer (0·1 M Tris-HCl, pH 7·0) to form the enzyme source. Aldolase activity was assayed by the method of Sibley and Lehninger[7], as modified by Beck[8].

3. Results and discussion

It is seen from Fig. 1, that in medium III, where only galactose is present as the carbon source, there is a pronounced decrease in the enzyme activity in all the mutant strains ; gal_5 and gal_9 record significantly low activities. In medium II containing galactose and glucose (3 : 1), gal_5 and gal_9 are grossly affected ; the activity of the enzyme is lowered in the other mutant strains too. But the decrease in activity in this medium is smaller when compared to the decrease in medium III. In a medium where glucose and galactose are present in equal proportions (Medium I), a slight decrease in enzyme activity is seen in the mutant strains, with gal_5 and gal_9 recording maximal changes. The activity of the enzyme in the various strains in a medium containing only glucose is shown in Fig. 1, Part IV.

It is thus seen that utilisation of galactose reduces aldolase activity in *gal* mutants. The extent of damage depends both on the nature of the medium as well as on the nature of the site of the mutation. The total mutants gal_5 and gal_9 are maximally affected in all the media. The slow-growing mutant (gal_9) and the partial mutant (gal_{10}) are affected to a lesser extent. As the concentration of galactose in the medium increases, the inhibitory effect on the enzyme activity also increases. The presence of glucose along with galactose appears to alleviate the toxic effects of galactose on these mutants.

The accumulation of galactose and/or some of its metabolic products intracellularly has been shown to inhibit several metabolic processes[9]. Aldolase has been shown to be specifically inhibited by galactose-1-PO_4[10] and this has been reported to accumulate in cells unable to utilise galactose[11]. The observed effect may also be attributed to an increased synthesis of aldolase by glucose derived metabolites. These factors either singly or in combination may account for the observed results.

The activity of the enzyme aldolase is a measure of the extent to which glycolysis is proceeding in a given tissue[12]. Glycolysis has been shown to be impaired in a variety of tissues subjected to galactose toxicity[13–16]. These reports support our observation that glycolytic activity as indicated by the activity of aldolase is affected in the 'galactosemic state'.

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