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SHORT REPORTS

SULPHITE STIMULATION OF BEAN MITOCHONDRIAL ADENOSINE TRIPHOSPHATASE

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INTRODUCTION

The inhibition of ATP formation in plant mitochondria by sulphite has been reported [1]. A possible explanation for the inhibition could be the sulphite stimulation of mitochondrial ATPase (EC 3.6.1.3). The stimulation of one form of yeast mitochondrial ATPase by 4 mM Na_2SO_3 has been reported, as well as the inhibition of yeast mitochondrial ATPase by 40 mM $(NH_4)_2SO_4$ [2]. Our purpose was to determine the influence of sulphite on bean mitochondrial ATPase in concentrations that inhibited ATP formation in bean mitochondria previously investigated [1].

RESULTS AND DISCUSSION

The K_m of the bean mitochondrial ATPase was 1.37 mM ATP and V_{max} was estimated as 322 nmol Pi/mg protein/min. Sulphite stimulated ATPase activity in concentrations that inhibited ATP formation in bean hypocotyl mitochondria (Table 1). Sulphate was apparently ineffective on ATPase when applied at 10 mM, a concentration in which sulphite stimulated ATPase. Sulphate had little effect on ATP formation at this concentration [1].

These data indicate that one possible mode of action of sulphite in inhibiting ATP formation in bean mitochondria is by stimulating ATPase. If the pollutant sulphur dioxide becomes sulphite as it enters plant cells, such a response could play a role in understanding the response of plants to sulphur dioxide pollution.

Earlier data demonstrated an inhibition of ATPase by sulphate [2] but $(NH_4)_2SO_4$ was employed rather than Na_2SO_4 and the concentration was 40 mM rather than 10 mM. Earlier studies have demonstrated that sulphite may stimulate one form of yeast mitochondrial ATPase but may inhibit two other forms of yeast mitochondrial ATPase [2].

Table 1. Effect of Na_2SO_3 and Na_2SO_4 on ATPase activity of bean hypocotyl mitochondria

Salt	Concentration (mM)	ATPase activity as % of control
Na_2SO_3	1	121
Na_2SO_3	3	137
Na_2SO_3	5	137
Na_2SO_3	7	152
Na_2SO_3	10	151
Na_2SO_3	15	186
Na_2SO_4	10	97

EXPERIMENTAL

Mitochondrial preparations were made from the etiolated hypocotyls of week-old etiolated bean seedlings (*Phaseolus vulgaris* cv Tendergreen Improved) as previously reported [1]. ATPase activity was determined by methods adapted from ref. [3]. The ATPase reaction was run at 30° in a mixture of the following in a final vol. of 5 ml: 0.1 M sucrose, 3.0 mM $MgCl_2$, 15 mM KCl, 1.37 mM ATP, 0.1 M tricine (pH 7.9). The equilibrium of bisulphite/sulphite at pH 7.9 is 1:9.8 [4]. The reaction was initiated by the addition of mitochondrial suspension containing 0.1 mg protein. Aliquots (1 ml) were withdrawn after 10 min and added to 2 ml ice-cold trichloroacetic acid. Pi was determined by the method of ref. [5]. Protein was determined by the method of ref. [6]. All determinations were replicated 4 × and involved four separate mitochondrial preparations.

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