

## ARTICLES

# INHIBITION OF SWEET POTATO $\beta$ -AMYLASE BY A POLYCATION

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### ABSTRACT

A novel polycation was synthesized. Inhibition of partially purified sweet potato  $\beta$ -amylase by the polycation was studied. Fifty per cent inhibition was obtained at 6.5 mM. Kinetic studies indicate that inhibition was noncompetitive and reversible, and  $K_i$  was 7.3 mM. The Hill coefficient was 1, suggesting that the polycation interacts with the enzyme non-cooperatively. Thermodynamic studies on enzyme-polycation complex indicate that the reaction is exothermic. The negative value of  $\Delta S$  indicates that the enzyme molecule undergoes conformational change during interaction with the polycation. The polycation also inhibits  $\beta$ -amylase derived from maize, wheat, barley and pea.

### INTRODUCTION

$\beta$ -AMYLASE ( $\alpha$ , 1-4 glucan maltohydrolase EC 3.2.1.2) catalyses the liberation of maltose from nonreducing ends of  $\alpha$ , 1-4 glucan. The interaction of enzyme with substrates, products and inhibitors has been investigated by kinetic and spectroscopic methods<sup>1</sup>. Inhibition of soybean  $\beta$ -amylase by glucose<sup>2</sup>, maltose<sup>3</sup> and cyclohexaamylase<sup>4</sup> have been demonstrated by kinetic studies.

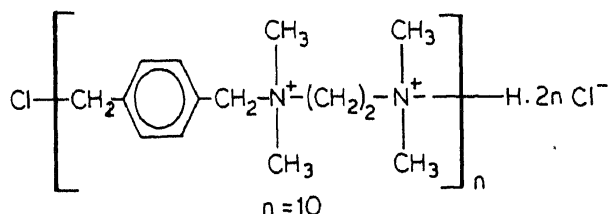
Biologically important polyamines such as putrescine, spermidine and spermine are cationic substances, and have been shown to interact with DNA and RNA and to affect the activities of several enzymes<sup>5-8</sup>. It was reported that synthetic polycations also strongly interact with nucleic acids<sup>9</sup>. Recent studies with a polycation showed that it was a potential inhibitor of mammalian amylases<sup>10,11</sup>. The present investigation was undertaken to study the inhibitory action of a synthetic polycation on partially purified sweet potato  $\beta$ -amylase.

### MATERIALS AND METHODS

Sweet potato  $\beta$ -amylase was purified according to the procedure of Hegde *et al.*<sup>12</sup> The enzyme from wheat, maize, barley and peanut was extracted according to Sharma *et al.*<sup>13</sup>

$\alpha, \alpha'$ -Dichloro-*p*-xylene (XDC) was purchased from AG Switzerland; *N,N,N',N'*-tetramethylethylenediamine (TEMED) from BDH, England; and soluble

starch and dimethyl formamide (DMF) from BDH, Bombay. The polycation shown below was synthesized according to the procedure of Rembaum and coworkers.<sup>14,15</sup>



The polycation was homogeneous. All other chemicals used were of analytical grade.

#### Enzyme assay

Enzyme activity was measured according to the method of Bernfeld<sup>16</sup> using dialysed starch as substrate. The amylase activity is expressed in terms of micromoles of reducing sugar liberated by 1 ml of enzyme per minute at room temperature. Protein concentration was determined according to Lowry *et al.*<sup>17</sup> using bovine serum albumin as standard.

#### Effect of polycation on plant amylases

Table 1 shows that the polycation inhibits  $\beta$ -amylase from maize, wheat, barley and pea to different extents in a concentration-dependent manner.

Detailed inhibition studies were carried out with sweet potato  $\beta$ -amylase. Different concentrations of polycation with 0.5 ml of enzyme and 0.5 ml of 1%