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Purification and biochemical characterization of a β-cyanoalanine synthase expressed in germinating seeds of *Sorghum bicolor (L.) Monech*

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Objective: The objective of this study is to purify β-cyanoalanine synthase from germinating seeds of sorghum to electrophoretic homogeneity and then determine its biochemical and catalytic properties.

Methodology: β-cyanoalanine synthase was isolated from sorghum seeds, purified using chromatographic techniques and its biochemical and catalytic properties determined.

Result: The purified enzyme had a yield of 61.74% and specific activity of 577.50 nmol H2S/min/mg of protein. The apparent and subunit molecular weight were 58.26±2.41 kDa and 63.4 kDa. The kinetic parameters with sodium cyanide as substrate were 0.67±0.08 mM, 17.60±0.50 nmol H2S/ml/min, 2.97x10-1 s-1 and 4.43x102 M-1s-1 for KM, Vmax, kcat and kcat/KM respectively. With L-cysteine as substrate, the kinetic parameters are 2.64±0.37 mM, 63.41±4.04 nmol H2S/ml/min, 10.71x10-1 s-1 and 4.06x102 M-1s-1. The optimum temperature and pH for activity were 35^{0}C and pH 8.5 respectively. The activation energy obtained was 131.75 J/mol/K. The enzyme retained more than half of its activity at 40^{0}C. Both monovalent and divalent ions enhanced enzyme activity. Inhibitors such as HgCl2, EDTA, glycine and iodoacetamide reduced enzyme activity.

Conclusion: The biochemical properties of the purified β-cyanoalanine synthase in germinating sorghum seeds highlight its roles in maintaining cyanide homeostasis.