O-ACETYLSERINE SULPHHYDROLASE FROM Cicer arietinum
PURIFICATION, PROPERTIES AND ITS ROLE IN AZIDE ACTIVATION INTO A MUTAGENIC METABOLITE.

by

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O-ACETYL SERINE SULFHYDRYLASE FROM
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ABSTRACT

O-acetylserine sulfhydrylase isolated from seven days old seedlings of chick peas (Cicer arietenum), was purified to apparent homogeniety by ammonium sulfate precipitation, gel filtration, affinity chromatography and non-denaturing gel electrophoresis. The purified enzyme was able to produce a mutagenic product from azide and O-acetylserine which increases the frequency of histidine reversions in Salmonella typhimurium TA1530 strain. The kinetic data presented in this study provide further supportive evidence for the role of O-acetylserine sulfhydrylase from Cicer arietenum in the conversion of azide to a mutagenic metabolite. The isolated enzyme has identical Km and Ki values for azide (N₃⁻). This strongly suggests that azide and the natural substrate (S⁻⁻) use the same catalytic site on the enzyme and demonstrates the role of O-acetylserine sulfhydrylase in the synthesis of azide mutagenic metabolite in Cicer arietenum.
The optimum pH of O-acetylserine sulfhydrylase found to be 8.0 and the enzyme preparation was stable to heating for 10 min. at 50°C.

The molecular weight of O-acetylserine sulfhydrylase from Cicer arietenum was estimated by gel filtration on sepharose 6B column to be 106,000 dalton. The subunit molecular weight was determined by SDS electrophoresis to be 54,000 dalton. These results clearly indicate that the molecular weight of Cicer arietenum enzyme is 108,000 dalton and it consists of two identical subunits. The enzyme absorption spectrum as well as the effect of specific inhibitors revealed the presence of pyridoxal phosphate group.