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Short Communication

EVALUATION OF SOME FUNGI FOR L-ASPARAGINASE PRODUCTION

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ABSTRACT
Sixty six (66) fungal isolates were screened for L- asparaginase production under batch culture condition. Among them, 52 fungal isolates showed L-asparaginase activity in biomass when grown in glucose - asparagine medium at 30ºC at static condition for seven days. However seven fungal isolates among them showed highest enzyme activity ranged between 116-235 IU/10 mg protein. Further, an experiment was conducted for the effect of carbon and nitrogen sources on L – asparaginase activity of selected fungal isolate T-22. The organism exhibited good L-asparaginase activity when grown in presence of fructose and proline. This is a preliminary study and more experiments on nutritional and cultural amendments for enhancement in enzyme production are needed for the bioprocess development.

INTRODUCTION
L-asparagine is an essential amino acid required for the growth of tumour cells whereas the growth of normal cells is independent of its requirement (Crowther, 1971; Mc Creadie *et al*., 1973; Swain *et al*., 1993). Wide range of bacteria, fungi, yeast, actinomycetes, algae, plants are found to be efficient producers of L-asparaginase (Savitri *et al*., 2003). Microbes are the better source of L-asparaginase, because they can be cultured easily and the extraction and the purification of L-asparaginase from them are also convenient, facilitating the large scale production. The fungal L- asparaginases have less adverse effects than bacterial L-asparaginase that causes an allergic reaction like skin rash, difficulty in breathing, decreased blood pressure, sweating or loss of consciousness. (Sarquis *et al*., 2004). In view to this, preliminary screenings of some fungi have been done under submerged culture condition.

***Key Words**: Enzymes, L-Asparaginase, Screening, Fungi and Optimization*

MATERIALS AND METHODS
Seven days old fungal cultures developed in glucose-asparagine was used for the enzyme extraction and analysis. The culture flasks were filtered to separate the fungal mat. The air dried biomass was taken as the source of enzyme. The biomass was completely homogenized with sterilized sand and normal saline in 1:10 ratio. It was centrifuged at 3000rpm for about 20 minutes The supernatant was used for the estimation of enzyme and protein (Soni , 1989, Bradford,1976). A potent isolate was further selected for optimization of carbon and nitrogen. A total number of 8 carbon sources i.e. Sucrose, Starch, Maltose, Lactose, Fructose, Raffinose and Mannitol were used in the broth keeping glucose as control. Like that 10 nitrogen sources i.e., L-Threonine, L-Proline, L-Tryptophan, L-Valine, L-Methionine, Aspartic acid, Glutamic acid, Xanthine and Yeast extract were used in the medium along with L-asparagine. The experiments were further carried with different % of fructose and L-Threonine, L-Proline and Aspartic acid. The method of harvesting, estimation of enzyme and protein was same through out the experiment.

RESULTS AND DISCUSSION

A total number of 66 fungal isolates were screened for L-asparaginase production from their biomass grown in glucose-asparagine broth medium at 30°C for seven days statically. Most of the fungal isolates have found to be positive for L- asparaginase production during screening process (Saxena and Sinha, 1981) . Among them, seven isolates showed higher enzyme activity ranged between 116-235 IU/10 mg protein. The fungal isolate T22 showed 235.0 IU/10 mg of protein when grown in presence of 1 % fructose and 0.5 % proline There was no enzyme activity recorded from the isolate cultured in the medium containing starch and mannitol as carbon source. Similarly, the fungal isolate didn't show any enzyme activity in presence of L-aspartic acid along with 1 % fructose. As this fungal isolate exhibited higher L – asparaginase activity, further experimentation is needed on nutritional and cultural amendments in order to confirm its potential for drug development.

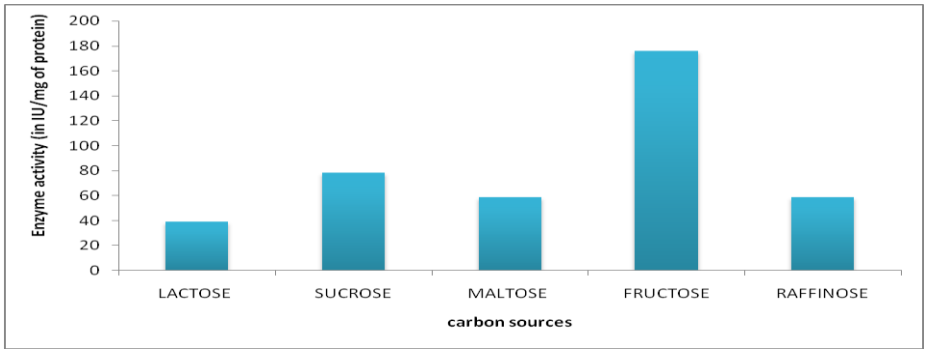

Figure 1. Effect of different carbon sources on L – asparaginase activity (IU/10mg protein)

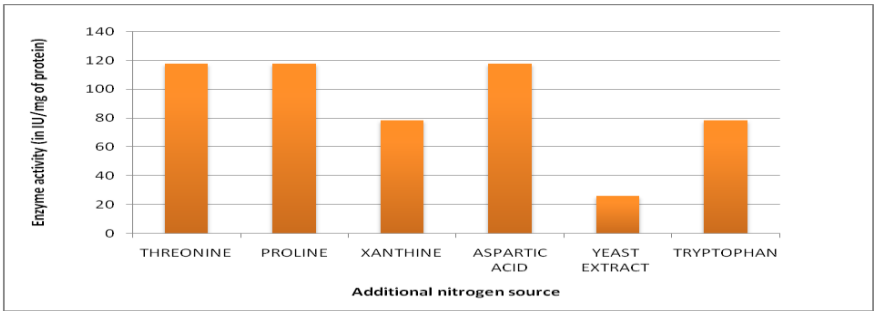

Figure 2. Effect of different nitrogen sources on L-asparaginase activity (IU/10 mg protein)

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