

## MICROBIAL PRODUCTION OF L-ASPARAGINASE PRODUCTION FROM SOIL MICROBE AND ITS MEDIUM OPTIMIZATION FOR BIOREMEDIATION OF AGRICULTURAL WASTE

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### ABSTRACT

Isolation of *E.coli* for the Production of L-asparaginase enzyme from bioremediating agricultural waste . From various microorganisms , L-asparaginase enzyme can be produced . These enzymes are helpful in the treatment of leukemia and it is also used in starchy food industries . From soil sample screening of L-asparaginase which is useful for producing bacteria was done . The screening is carried by two methods ie, primary screening and secondary screening. The primary screening is done by qualitative method .here rapid plate assay is used as a qualitative method the secondary screening is done by nesslerization method. The bacterial isolates A6, c11, C19 . Were bacillus species and all were staphylococcus species which is observed pink color zone in primary screening . It was identified based on morphological , cultured and biochemical test. L-asparaginase enzyme that used to make proteins and create a new cells as a medication and in food manufacturing . It is approved for medical use in the United states from 1978 onwards.

**Keywords :** *E.coli*, L. Asparaginase, Agricultural waste, Bacillus species, Bacterial isolates.

### 1. INTRODUCTION

*Escherichia coli* L. Asparaginase anti-tumor activity was first established by Broome (1961) and Mashburn and Wriston(1964) , it's production utilizing microbial system considered as cost effective and eco-friendly nature. L. Asparaginase is a class of enzymes that hydrolyzes the aminoacids L. Asparaginase into aspartic acid and ammonia. The enzyme is also known to have an allosteric site, which can be bound by an effector molecule to alter its activity. L. Asparaginase is an amidase enzyme, it's catalyzes the conversion of amino acid L-asparagine to L-aspartate and ammonia [13]. The amino acid L-asparagine be in amide group is non essential aminoacid and nutritional requirement of both normal and cancerous cells.

#### Catalytic reaction of L-asparaginase

Hydrolysis of L-asparagine to L-aspartic acid. Since leukemia cells are unable to produce its own endogenous L-asparagine in a sufficient amount, the reduction of blood circulating L-asparagine induce them to starve without affecting healthy cell.

### 2. SOURCES OF L-ASPARAGINASE ENZYME AND THEIR TYPES

#### Sources

L-asparaginase formulations are derived naturally from bacterial sources like *Escherichia coli* and *Erwinia chrysanthemi* . There are many number of sources available for isolation of L-asparaginase enzyme, they are bacterial, fungal, plant, animal sources.

#### Plant sources

The roots of *Pinus pinaster* and *Pinus radiata* has an activity of L.Asparaginase in soil. On the basis of high specificity, *Withania somnifera* is a conceivable source of L. Asparaginase enzyme.

#### Fungal source

By the filamentous fungi like *Aspergillus tamarii* and *Aspergillus terreus* , the enzyme L. Asparaginase is produced. The best nitrogen source for this enzyme production from *A.terreus* is L.proline.

#### Bacterial source

*Pseudomonas aeruginosa* , *Pyrococcus furiosus*, *Thermus thermophilus* , *Serratia marcescens* had been found to be a good Asparaginase source for this enzyme production.

#### Animal source

L. Asparaginase enzyme is typically derived from *Escherichia coli*, a species of bacteria found in the gut of animals. In some cases, the enzyme can be derived from other sources, such as chicken embryo, porcine pancreas, or baker's yeast.

### Types of L.Asparaginase enzyme

There are 3 types of L-asparaginase enzyme. They are *Escherichia coli* which is also known by its brand spectrila, *Erwinia chrysanthemia* bacteria which is also known as Erwinase ,pagylated which is also known as pegaspargase.

## 3. APPLICATIONS OF L-ASPARAGINAS

### Pharmaceutical Industry : Antineoplastic Action

L.asparaginase is used as chemotherapeutic agent in the treatment of lymphoma disease and lymphoproliferative. It plays major role in chemotherapeutic protocols such as acute hodgkin's lymphomas and acute lymphoblastic leukemia. From blood serum , asparagine is hydrolyzes by l.asparaginase. for fast and malignant growth, cancer cells require high amount of asparagine.

### Anticancer Drug

For many tumor cells, l.asparaginase is essential amino acid with is used for protein synthesis and cell growth. L.asparaginase is deliberated as important antitumor drug . L.asparaginase is described widely in animals, plants and microorganisms . Combination of chemotherapeutic agent with other drugs is used it the treatment of malignancies.

### Food Industry: Acrylamide Formation

L.asparaginas is mainly responsible for acrylamide production.it is used in fried and backed foods.in 2003,zyzak et Al identified that the amide chain is provided from L.asparaginase Decreasing acrylamide quantity in foods cause estimated Strategies in reagent reduction or removed .

### Biosensor

To analyse the asparaginase level in leukemia , l.asparaginase is used for to develop biosensor. For l.asparaginase analysis, XRD,SEM and TEM are used a change in PH , colour and absorption due to Production of ammonium ion from the hydrolysis of asparaginase.

### Amino Acid Metabolism

L. Asparaginase plays a major role in biosynthesis of an aspartic family of amino acids, like lysine, threonine, methionine. According to Kreb's cycle , aspartic acid is direct progenitor of lysine , threonine and is formed by the action of L-asparaginase enzyme [164]

## 4. CONCLUSION

In this review article, we have presented the importance of L-asparaginase both in pharmaceutical and food industry. L-asparaginase , an amilohydrolase enzyme has wide range of applications. Though disseminate generally in plant, bacteria, animal and fungi, microbial system has considered as cost effective and easy production from microbes. More over, yields of L. asparaginase and gene expression rate of this enzyme is very low. Amino acids generally stimulated the production of enzyme L. asparaginase enzyme used in the treatment of human neoplastic disease which the enzyme be produced in large quantities.

## 5. REFERENCE

- [1] Hill J.M . L- asparaginase therapy for leukemia and other malignant neoplasms . J Am med Assoc. 1967 ; 202(9) :882.
- [2] Van den berg H.asparaginase revisited. Leuk Lymphoma . 2011;52(2) : 168-178.
- [3] Lang s.uber desamidierung im Tierkorper .Beitr chem physiol pathol . 1904; 5: 321-345.
- [4] Verma N., Kumar k., Karur G., Anand s. L-asparaginase : a promising chemotherapeutic agent. Crit Rev Biotechnol . 2007;(1) : 45-62.
- [5] Mohan Kumar N.s ., Shimray c.A ., Indrani D., Manonmani H.K. Reduction of acrylamide formation in sweet bread with L-asparaginase Treatment . Food Bioprocess Technol. 2013;7(3): 741 -748.
- [6] Keating M.J., Holmes R., Lerner s., Ho D.H. L-asparaginase and PEG asparaginase -past , present, and future. Leuk Lymphoma. 1993 ;10 (suppl) : 153 – 157.
- [7] Furth o., Fridemann M. Uber die verbreitung asparaginspaltender organfermente . Biochemistry . 1910; 26: 435 -440.
- [8] Broome J.D . Antilymphoma activity of L.asparaginase in vivo : clearence rates of Enzyme preparations from guinea pig serum and yeast in relation to their effect on tumor growth. J Natl cancer Inst. 1965;35(6):967-974.
- [9] Apple I.M., van kessel-Bakvis c., Stigter R., Pieters R. Influence of two different regimers of concomitant treatment with asparaginase and dexamethasone on hemostasis in childhood acute lymphoblastic leukaemia. Leukemia. 2007; 21(11) :2377 – 2380.

- [10] Mmedeiros vinci R., Mestdagh F., De meulenaer B. Acrylamide formation in fried potato products -present and future , a critical review on mitigation strategies. Food chem. 2012 ;133(4):1138-1154.
- [11] Prakasham, RS, subha Rao , CH,2007, L-asparaginase Production by isolated staphylococcus sp. -6A: design of experiment considering interaction effect for process parameter optimization, journal of applied microbiology, 102(5)1382-1391.
- [12] Soni yadav, Sitansu kumar varma, Jitendra Singh,ajay kumar, 2014, industrial Productions and clinical applications of L-asparaginase: a chemotherapeutic agent, international journal of biotechnology and bioengineering 8(1),54-60.
- [13] Moorthy vidhya, Ramalingam Aishwarya, 2010, Production, purification, characterization of extracellular L-asparaginase from soil Isolate of bacillus sp., African journal of microbiology research 4(18),1862-1867.
- [14] Alapati kavitha, Muvva Vijaylakshmi, 2012, a syudy on L-asparaginase of nocardia levis Mk-VL 113, The scientific World journal 2012.
- [15] Maurer, H.R. (2001). Bromelain ; biochemistry pharmacology and medical use. Cellular and molecular life sciences, 58, 1234-1245.
- [16] Hammes , G.G.(2008).how to Enzymes really work journal of biological chemistry 283, 22337- 22346.
- [17] Kumar, k., & Verma,N.(2012) . The various sources and application of L-asparaginase.asian journal of biochemical and pharmaceutical research,3,197-205.
- [18] Lang, s., &Uber. (1904). Desamidierung im Tierkorper. Betraege zur chemischen physiologic and pathologie, 190 , 5321-345.
- [19] Clementi , A. (1992), la desamidation enzymatique de L-asparaginase chez les differentes especes animals et la signification physiologique de sa presence dans l'organiasma . Archives internationalex de physiologic, 19, 369- 376.
- [20] Broome , J.D. (1961), evidence that the L-asparaginase activity of guinea pig serum is responsible for its anti-lymphoma effects. Nature, 191,1114-1115.
- [21] Broome,J.D. (1968) , factors which may be influence the effectiveness of L-asparaginase as tumor inhibitors. British journal of cancer,22,595-602.
- [22] Clubb, j.s., neale, f.c ., & Posen, s. (1965) . The behaviour of infused human placental alkaline phosphatase in human subjects, journal of laboratory and clinical medicine,66,493-507.
- [23] Mecoy, T.A., Maxwell, M., & Neuman, R.E.(1956). The amino acid requirements of the Walker carcino sareoma 256 in vitro cancer research, 16,979-984.
- [24] Altenbern, R.A., & housewright,R.D. (1954) , stereo specific asparaginase in smooth brucella abortus strain 19. Archives of biochemistry and biophysics, 49,130-137.
- [25] Holmquist, J.D, (1963) . Effect of normal Sera of several related rodents on 6C3HED lymphoma in Vivo.experimental biology and medicine, 113,444-445.
- [26] Broome, J.D . (1963) . Evidence that the L-asparaginase of guinea pig serum is responsible for its anti-lymphoma effects 11 lymphoma 6C3HED cells cultured in a medium devoid of L-asparaginase lose their susceptibility to the effect of guinea pig serum in vivo. Journal of experimental medicine, 118, 121-148.
- [27] Ohnuma, T., Bergel, F., & Bray, R.c,(1967). Enzyme in cancer asparaginase from chicken liver. Biochemical journal, 103,238-245.
- [28] Campbell, H.A., mashburn, L. T., Boyse, E.A., & Old, L.J.(1967). Two L-asparaginase from Escherichia coli B. Their separation purification and antitumor activity, biochemistry;6,721-730.
- [29] Kotzia, G.A., & labrou, N,E. (2007). L-asparaginase from Erwinia chrysanthemi 3937; cloning expression and characterization., Journal biotechnology; 127,657-669.
- [30] Savitri, N.A., & azrni, W. (2003). Microbial L-asparaginase: a potent antitumor Enzyme. Indian journal of biotechnology;2,184-194.