iJRASET
International Journal For Research in Applied Science and Engineering Technology

INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Volume: 6 **Issue:** II **Month of publication:** February 2018

DOI:

www.ijraset.com

Call: 08813907089 | E-mail ID: ijraset@gmail.com

International Journal for Research in Applied Science & Engineering Technology (IJRASET)
ISSN: 2321-9653; IC Value: 45.98; SJ Impact Factor: 6.887
Volume 6 Issue II, February 2018- Available at www.ijraset.com

Screening of L-Asparaginase Producing Bacteria from Central MP

Deepali Shukla[1], Vivek Kumar Shrivastav[2], Dr. Archana Shrivastav[3]
[1, 2]*Department of microbiology, college of Life sciences Cancer hospital and research institute,Gwalior MP*
[3]*Prof. And HeadDepartment of microbiology, college of Life sciences Cancer hospital and research*

Abstract: L-Asparaginase is devlop a remarkable achievement in the field of medicine as an effective antitumor agent Asparaginase has proved to be promising for the treatment of acute lymphocytic leukemia. L-Asparaginase is the most important agent used in multi-drug chemotherapy regimens in the treatment of malignancies which are derived from the lymphoid system (Acute Lymphoblastic leukemia and Non-Hodgkin lymphoma).Fact behind this property of enzyme is that, the malignant cell deficient in aspartate-ammonia ligase activity, which restricts their ability to synthesize normally non-essential amino acid, L-asparagine, which requires for growth and therefore, they are forced to extract it from body fluids. The action of Asparaginase does not affect the functioning of normal cells, which are able to synthesize enough for their own requirements, but reduce the free exogenous concentration, and so induce a state of fatal starvation in the susceptible tumor cells. The present deals new search of potential L-Asparaginase from bacteria for screening of L-asparaginase producer bacteria from soil of Madhya Pradesh.

Keywords: L-Asparaginase, ALL, Non-Hodgkin lymphoma

I. INTRODUCTION

L- Asparagianse is an antileukamic enzyme, having promising properties for the treatment of acute lymphocytic leukaemia[1], it is proved by the research. Fact behind this property of enzyme is that, the malignant cell deficient in aspartate-ammonia ligase activity, which restricts their ability to synthesize normally non-essential amino acid, L-asparagine, which requires for growth and therefore, they are forced to extract it from body fluids. The action of Asparaginase does not affect the functioning of normal cells, which are able to synthesize enough for their own requirements, but reduce the free exogenous concentration, and so induce a state of fatal starvation in the susceptible tumor cells. Emergence of L-asparaginase as a potential health care agent for the treatment of acute lymphocytic leukemia [2],[3],[4]and[5] and increasing demand of L-asparaginase in food industries have rise great attention to scientific communities and research industries. With the development of its new functions, a great demand for L-asparaginase is expected in coming years, L-asparaginase production using microbial systems has attracted considerable attention. Bacteria are promising source of a wide range of important enzymes with respect to their medicinal use, some of which produced in industrial scale. The present work initiates to address new search of potential L-Asparaginase from bacteria for fulfilling this aim. Present study deals screening of L-asparaginase producer soil bacteria of Madhya Pradesh. Madhya Pradesh represents central India. Soil of MP has diverse of temperature, pressure, pH, salinity and water activity at which environment with greater microbial diversity might claim the production of enzyme with some properties that can stabiles at different temperature tolerance. A systematic study to explore the soil sources for bacteria producing L-asparaginase, was carried out. The study included identification of the isolate; optimization of process parameters; purification and characterization of the enzyme.

II. METHODS AND MATERIAL

A. Sample Collection

The study was conducted from June2010 to August 2012.Sediment samples were collected from different soil in the sterilized poly bags from the depth of 30cm of different farms in the various area of Madhya Pradesh in summer .

B. Isolation of Bacteria

Isolation of bacteria from soil was done by conventional method [**6**]. Isolation was done by taking 1 gm of each soil sample to 100 ml of sterile water. The suspension was kept on a rotary shaker for 30 min. and allows settling the suspending matter. One ml of the supernatant was serially diluted with sterile water. 20 ml sterile nutrient agar added to one ml of each dilution mixed thoroughly and

plated in 9 cm diameter sterile petri-dishes and incubated at 37ºC for 24 hrs. Fluconazole-75 µg/ml was incorporated to prevent the fungal contamination.

C. Screening

1) *Rapid plate Semi Quantitative Assay: Isolates* were initially screened by rapid plate assay method, using modified M-9 medium **[7]**. Isolated colonies from nutrient agar were inoculated on specific medium for primary screening. After inoculation, plates were then incubated at 37°C for 24-48 hrs. A set of plates was also run as a control without L-Asparagine. The strains having potential for L-Asparaginase production were selected on the basis of pink colour zone formation around the colonies on M9 medium and were used for further screening.

D. Secondary Screening

1) *Inoculum Preparation:* Quantitative estimations of enzyme activities were carried out using liquid M-9 medium **[7]** Each isolated bacterial colony from the primary screening were inoculated in liquid M-9 medium at pH 7.0 and incubated at 37ºC at 120 rpm for 24 to 72 h, in a controlled environment shaker-incubator. Un-inoculated medium served as a control. After incubation, the culture broths were centrifuged at 10,000 ×g for 10 min, and the supernatant were collected for enzyme assay.

E. Analytical Study

1) *Determination of L-Asparaginase activity :*Determination and analysis of L-Asparaginase activity was done according to the standard protocol of Imada*et al.,* 1971[8].

F. Protein Estimation.

Extracellular protein was estimated by Lowry et al., 1951[9].

III. RESULTS

A. Isolation&Primary screening of L-Asparaginase producing bacteria

The present investigation was targeted to screen the potentiality of L-Asparaginase production by soil bacteria, 70 different soil samples were collected from various locations of Madhya Pradesh. Out of 70 samples 20, 10,10,10,10 were collected from Gwalior, Banmore, Bhind, Dabra, and Morena region respectively. A total 312 bacteria were isolated from 70 samples. Out of them 229 were found positive for L-Asparaginse activity based on primary screening. Table 1 reveals that from total isolates of 312, 75 bacteria were isolated from Gwalior, 55 from Banmore, 56 from Bhind, 67 from Dabra, and 59 bacteria were from Morena region.

Out of 75 isolates from Gwalior region 63 (84%) isolates were found to be L-Asparaginase producers, 23 (41.8%) isolates out of 55 isolates from Banmore region were positive for L-Asparaginase activity similarly, out of 56, 47(83.9%) isolates were having positive L-Asparaginase activity in Bhind and from Morena region 41 (74.5%) out of 59 isolates shows L-Asparaginase activity. Data from table I reveals that on primary screening overall 229(73.39%) out of 312 of bacteria were found to have potential for L-Asparaginse production isolated from soil of Madhya Pradesh. Primary screening for L-Asparaginase production shown in. Figure 2.

Table 1: Primary screening of L-Asparaginase producing isolates from various region of Madhya Pradesh

Region	Soil Sample	Total Isolates	L-Asparaginase producing Isolates	
			Number	Percentage
Gwalior	20	75	63	84%
Banmore	10	55	23	41.8%
Bhind	10	56	47	83.9%
Dabra	10	67	55	83.3%
Morena	10	59	41	74.5%
Total	70	312	229	73.39%

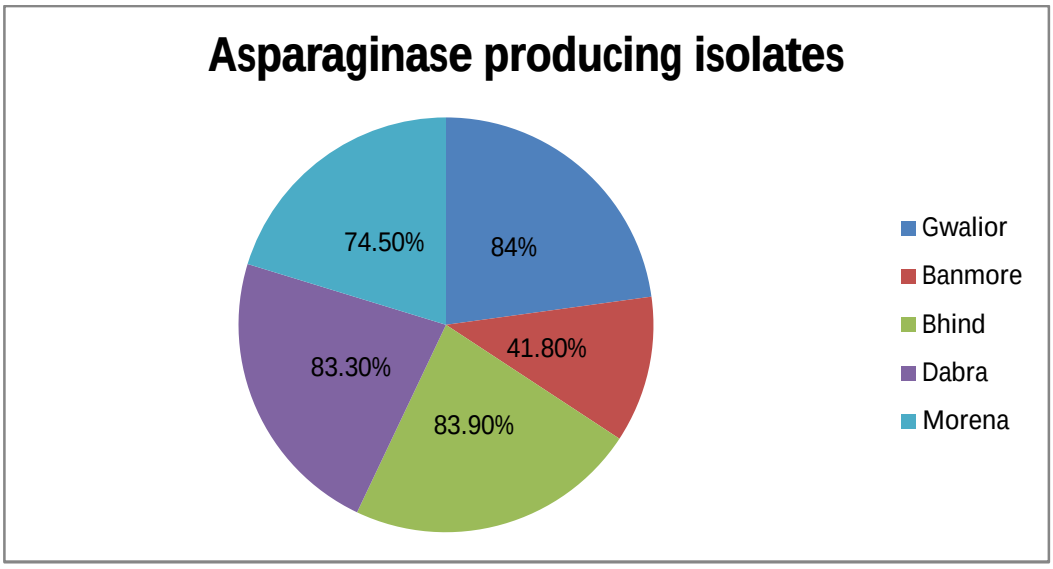


Fig1: L-Asparaginase producing isolates from various region of Madhya Pradesh

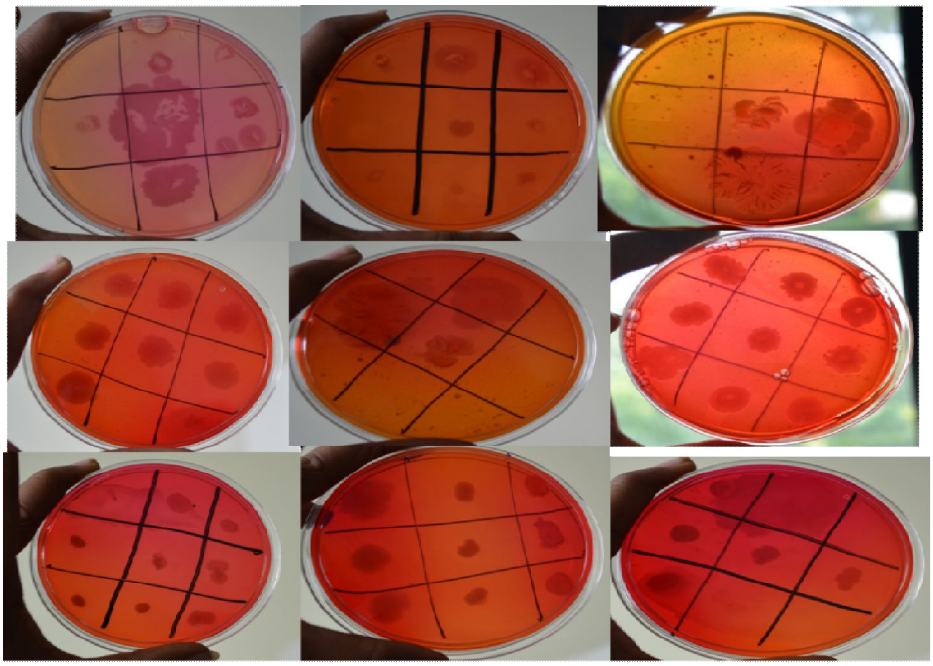


Figure 2: Evaluation of L-Asparaginase production capability by bacteria on primary screening

B. Secondary Screening

After primary screening, L-Asparaginase produced by isolates was quantitatively estimated with the help of secondary screening by the method of Nesselerisation. after secondary screening, all 229 isolate were selected as primary screening ranked into four categories for enzyme activity, poor L-Asparaginase producers, moderate L-Asparaginase producers, good L-Asparaginase producers and excellent, L-Asparaginase producer. (*Catagories=Less than 0.1 IU/ml =Poor L-Asparaginase producers,0.1 IU/ml-0.5 IU/ml= Morderate L-Asparaginase producers,0.5 IU/ml-1 IU/ml= Good L-Asparaginase producers, Greater than 1 IU/ml = Excellent L-Asparaginase producers)Region wise study indicates, that from Gwalior region, 63(84%) isolates were found to be positive for L-Asparaginase activity after primary screening, secondary screening reveals that maximum 29 (46%) isolates were found to be poor L-Asparaginase producer, 18 (28.5%) shows moderate L-Asparaginase activity, 10(15.8%) were found to be good, and 6 (9.5%) isolates were found to have excellent L-Asparaginase activity, Table 2 present the data of L-Asparaginase activity of bacteria isolated from Gwalior.

Table 2: L-Asparaginase activity of isolates from Gwalior region

S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation	S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation
1	CLS01	1.37	1.067	42	CLS42	1.63	1.623333
2	CLS02	1.094	1.113333	43	CLS43	1.752	1.733
3	CLS03	1.64	1.6533	44	CLS44	0.355	0.355333
4	CLS04	0.05	0.050667	45	CLS45	0.504	0.496
5	CLS05	0.178	0.129333	46	CLS46	0.436	0.441
6	CLS06	0.69	0.65	47	CLS47	0.62	0.618333
7	CLS07	0.082	0.074	48	CLS48	0.505	0.585333
8	CLS08	0.353	0.352333	49	CLS49	0.374	0.376667
9	CLS09	1	1.02	50	CLS50	1.413	1.402667
10	CLS10	0.777	0.755	51	CLS51	0.58	0.586333
11	CLS11	0.735	0.743667	52	CLS52	0.588	0.593
12	CLS12	1.14	1.153333	53	CLS53	0.516	0.509333
13	CLS13	0.8	0.792333	54	CLS54	0.577	0.593
14	CLS14	0.773	0.766	55	CLS55	0.338	0.332333
15	CLS15	0.742	0.743	56	CLS56	1.63	1.623333
16	CLS16	0.277	0.264	57	CLS57	0.303	0.304667
17	CLS17	0.065	0.105667	40	CLS40	0.056	0.063
18	CLS18	0.076	0.086333	41	CLS41	1.18	1.17
19	CLS19	1.055	1.049	42	CLS42	1.63	1.623333
20	CLS20	0.082	0.086667	43	CLS43	1.752	1.733
21	CLS21	1.31	1.357	44	CLS44	0.355	0.355333
22	CLS22	0.669	0.669667	45	CLS45	0.504	0.496
23	CLS23	0.985	0.995	46	CLS46	0.436	0.441
24	ClS24	0.669	0.667	47	CLS47	0.62	0.618333
25	CLS25	0.747	1.162667	48	CLS48	0.505	0.585333
26	CLS26	1.094	1.113333	49	CLS49	0.374	0.376667
27	CLS27	0.108	0.11	50	CLS50	1.413	1.402667
28	CLS28	0.225	0.225	51	CLS51	0.58	0.586333
29	CLS29	0.13	0.131667	52	CLS52	0.588	0.593
30	CLS30	0.439	0.440333	53	CLS53	0.516	0.509333
31	CLS31	0.502	0.53	54	CLS54	0.577	0.593
32	CLS32	0.367	0.342	55	CLS55	0.338	0.332333
33	CLS33	0.183	0.184667	56	CLS56	1.63	1.623333
34	CLS34	0.362	0.368	57	CLS57	0.303	0.304667
35	CLS35	0.107	0.154	58	CLS58	0.226	0.227333
36	CLS36	1.18	1.15	59	CLS59	0.18	0.180333
37	CLS37	0.217	0.214667	60	CLS60	0.764	0.765333
38	CLS38	0.137	0.131333	61	CLS61	0.691	0.687667
39	CLS39	0.285	0.282333	62	CLS62	0.385	0.382667
40	CLS40	0.056	0.063	63	CLS63	0.421	0.417667
41	CLS41	1.18	1.17				

In Banmore region, a total of 23 isolates were found L-Asparaginase producers. Quantitative estimation indicates that from Banmore region 7 (30.43%) isolates were found poor L-Asparaginase producer, maximum isolates 9(39.1%) shows moderate L-Asparaginase activity, 3(13%) were found good, and 4(17.3%) isolates were found to have excellent L-Asparaginase activity, Table 3 shows results of enzyme activity.

Table 3: L-Asparaginase activity of isolates from Banmore region

S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation	S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation
1	CLS164	0.035	0.057333	12	CLS175	1.31	1.357
2	CLS165	0.09	0.091	13	CLS176	0.669	0.669667
3	CLS166	0.126	0.13	14	CLS177	1.29	1.266667
4	CLS167	0.061	0.060333	15	CLS178	0.747	1.162667
5	CLS168	0.101	0.112333	16	CLS179	1.094	1.113333
6	CLS169	0.094	0.092333	17	CLS180	1.29	1.273333
7	CLS170	0.054	0.054667	18	CLS181	0.05	0.050667
8	CLS171	0.065	0.105667	19	CLS180	0.178	0.129333
9	CLS172	0.076	0.086333	20	CLS181	0.072	0.073333
10	CLS173	1.055	1.049	21	CLS182	1.29	1.343667
11	CLS174	0.082	0.086667	22	CLS183	0.074	0.072
				23	CLS184	0.184	0.172333

47 isolates was found to be positive L-Asparaginase producers after primary screening, from Bhind region 19 (40.4%) isolates were found to be poor L-Asparaginase producer, 15 (31.9%) show moderate L-Asparaginase Activity, 10 (21.2%) were found good, and 3(6.3%) isolates were found to have excellent L-Asparaginase activity Table 4 represents the enzyme activity result of bacteria isolated from Bhind region.

Table 4: L-Asparaginase activity of isolates from Bhind region

S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation	S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation
1	CLS64	0.383	0.377333	23	CLS87	0.082	0.074
2	CLS65	0.298	0.295667	24	CLS88	0.353	0.352333
3	CLS66	0.09	0.091	25	CLS89	0.356	0.358333
4	CLS67	0.126	0.13	26	CLS90	0.381	0.362
5	CLS68	0.061	0.060333	27	CLS91	0.0422	0.041
6	CLS69	0.101	0.112333	28	CLS92	0.156	0.159333
7	CLS70	0.094	0.092333	29	CLS93	0.8	0.792333
8	CLS71	0.054	0.054667	30	CLS94	0.773	0.766
9	CLS72	0.065	0.105667	31	CLS95	1.08	1.09
10	CLS73	0.126	0.13	32	CLS96	0.277	0.264
11	CLS74	0.076	0.086333	33	CLS97	0.24	0.217
12	CLS75	1.055	1.049	34	CLS98	0.102	0.109333
13	CLS76	0.082	0.086667	35	CLS99	0.116	0.116667
14	CLS78	1.31	1.357	36	CLS100	0.739	0.729
15	CLS79	1.33	1.33	37	CLS101	0.353	0.352333
16	CLS80	0.985	0.995	38	CLS102	0.356	0.358333
17	CLS81	0.747	1.162667	39	CLS103	0.381	0.362
18	CLS82	1.094	1.113333	40	CLS104	0.0422	0.041

19	CLS83	0.759	0.740333	41	CLS105	0.156	0.159333
20	CLS84	0.05	0.050667	42	CLS106	0.293	0.284667
21	CLS85	0.178	0.129333	43	CLS107	0.0413	0.044667
22	CLS86	0.072	0.073333	44	CLS108	0.05	0.044967

L-Asparaginase activity has been noted in 55 (83.3%) isolates from Dabra on primary screening, maximum isolates having poor L-Asparaginas activity that is 22 (40%), while 18 (32.7%),10 (18%),5(9%) isolates were moderate, good and excellent L-Asparaginase producers respectively (Table 5).

Table5: L-Asparaginase activity of isolates from Dabra region

S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation	S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation
1	CLS112	0.77	0.778333	27	CLS138	0.285	0.282333
2	CLS113	0.8	0.792333	28	CLS139	0.056	0.063
3	CLS114	0.773	0.766	29	CLS140	0.553	0.531667
4	CLS115	0.742	0.743	30	CLS141	0.639	0.636333
5	CLS116	0.277	0.264	31	CLS142	1.752	1.733
6	CLS117	0.24	0.217	32	CLS143	0.355	0.355333
7	CLS118	0.102	0.109333	33	CLS144	0.504	0.496
8	CLS119	0.116	0.116667	34	CLS145	0.436	0.441
9	CLS120	0.739	0.729	35	CLS146	0.62	0.618333
10	CLS121	0.571	0.575667	36	CLS147	0.505	0.585333
11	CLS122	0.086	0.084	37	CLS148	0.374	0.376667
12	CLS123	0.66	0.683333	38	CLS149	1.413	1.402667
13	CLS124	0.476	0.475	39	CLS150	0.58	0.586333
14	CLS125	0.439	0.440333	40	CLS151	0.588	0.593
15	CLS126	0.108	0.11	41	CLS152	0.516	0.509333
16	CLS127	0.225	0.225	42	CLS153	0.577	0.593
17	CLS128	0.13	0.131667	43	CLS154	0.338	0.332333
18	CLS129	0.439	0.440333	44	CLS155	0.276	0.280333
19	CLS130	0.502	0.53	45	CLS156	0.303	0.304667
20	CLS131	0.367	0.342	46	CLS157	0.226	0.227333
21	CLS132	0.183	0.184667	47	CLS158	0.18	0.180333
22	CLS133	0.362	0.368	48	CLS159	0.764	0.765333
23	CLS134	0.107	0.154	49	CLS160	0.691	0.687667
24	CLS135	0.502	0.53	50	CLS161	0.385	0.382667
25	CLS136	0.217	0.214667	51	CLS162	0.421	0.417667
26	CLS137	0.137	0.131333	52	CLS163	0.383	0.377333

In morena41 (74.5%) isolates have shown L-Asparaginase activity out of which 15 (36.5%) isolates were observed poor L-Asparaginase producer, 12(29.2%) show moderate L-Asparaginase activity, 7(17%) were observed good, and 7 (17%) isolates were observed to have excellent L-Asparaginase activity, (Table 7).

Table 7: L-Asparaginase activity of isolates from Morena region

S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation	S.No.	Isolate Number	Enzyme Activity IU/ml	Standard Deviation
1	CLS185	0.164	0.157	23	CLS208	0.088333	0.118778
2	CLS186	0.102	0.108333	24	CLS209	0.011333	0.084889
3	CLS187	0.11	0.109667	25	CLS210	0.038667	0.126444
4	CLS188	0.124	0.128667	26	CLS211	0.026667	0.090667
5	CLS189	0.045	0.045333	27	CLS212	0.1132	0.121511
6	CLS190	0.057	0.048667	28	CLS213	0.059	0.056444
7	CLS191	0.05	0.048333	29	CLS214	0.012333	0.046667
8	CLS192	0.156	0.126	30	CLS215	0.132	0.135333
9	CLS193	0.047	0.092333	31	CLS216	0.011	0.049
10	CLS194	0.069	0.071667	32	CLS217	0.051	0.053556
11	CLS195	0.079	0.079	33	CLS218	0.04	0.053333
12	CLS197	0.05	0.053333	34	CLS219	0.11	0.113333
13	CLS198	0.058	0.052333	35	CLS220	0.046	0.048667
14	CLS199	0.013	0.057889	36	CLS221	0.046	0.048667
15	CLS200	0.035	0.057333	37	CLS222	0.12	0.133333
16	CLS201	0.0598	0.060756	38	CLS223	0.089	0.087333
17	CLS202	0.010333	0.041889	39	CLS224	0.061	0.058667
18	CLS203	0.012667	0.050333	40	CLS225	0.08	0.1
19	CLS204	0.014667	0.057444	41	CLS226	0.091	0.093667
20	CLS205	0.040333	0.056333	42	CLS227	0.0922	0.093844
21	CLS206	0.031	0.123667	43	CLS228	0.099	0.094333
22	CLS207	0.0235	0.072722	44	CLS229	0.12	0.106667

C. Quantitative status of L-Asparaginase producing isolates

As data reveals that out of total 229 L-Asparaginase producing isolates 53 (23%) isolates were found poor L-Asparaginase producers, followed by 84 (36.6%) as moderate producers, 67(29.2%) as good producers and 25 (10%) were as excellent producers of L-Asparaginase (Fig 3,4 and Table 8).

Fig 3: Quantitative status of L-Asparaginase producing isolates in Madhya Pradesh

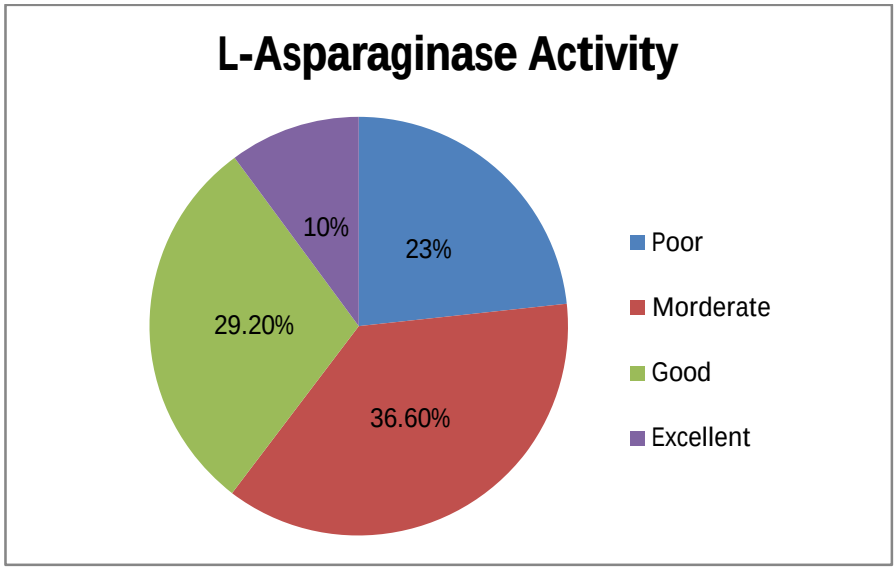


Table8: Quantitative status of L-Asparaginase producing isolates

Region	Number of Isolates	Catagories of L-Asparaginase Producing Isolates							
		Poor L-Asparaginase producers		Morderate L-Asparaginase producers		Good L-Asparaginase producers		Excellent L-Asparaginase producers	
		Number of Isolates	Percentage of Isolates	Number of Isolates	Percentage of Isolates	Number of Isolates	Percentage of Isolates	Number of Isolates	Percentage of Isolates
Gwalior	63	29	46%	18	28.5%	10	15.8%	6	9.5%
Banmore	23	7	30.43%	9	.39.1%	3	13%	4	17.3%
Bhind	47	19	40.4%	15	31.9%	10	21.2%	3	6.3%
Dabra	55	22	40%	18	32.7%	10	18%	5	9%
Morena	41	15	36.5%	12	29.2%	7	17%	7	17%
Total	229	53	23%	84	36.6%	67	29.2%	25	10 %

*Catagories=Less than 0.1 IU/ml =Poor L-Asparaginase producers
0.1 IU/ml-0.5 IU/ml= Morderate L-Asparaginase producers
0.5 IU/ml-1 IU/ml= Good L-Asparaginase producers
Greater than 1 IU/ml = Excellent L-Asparaginase producers

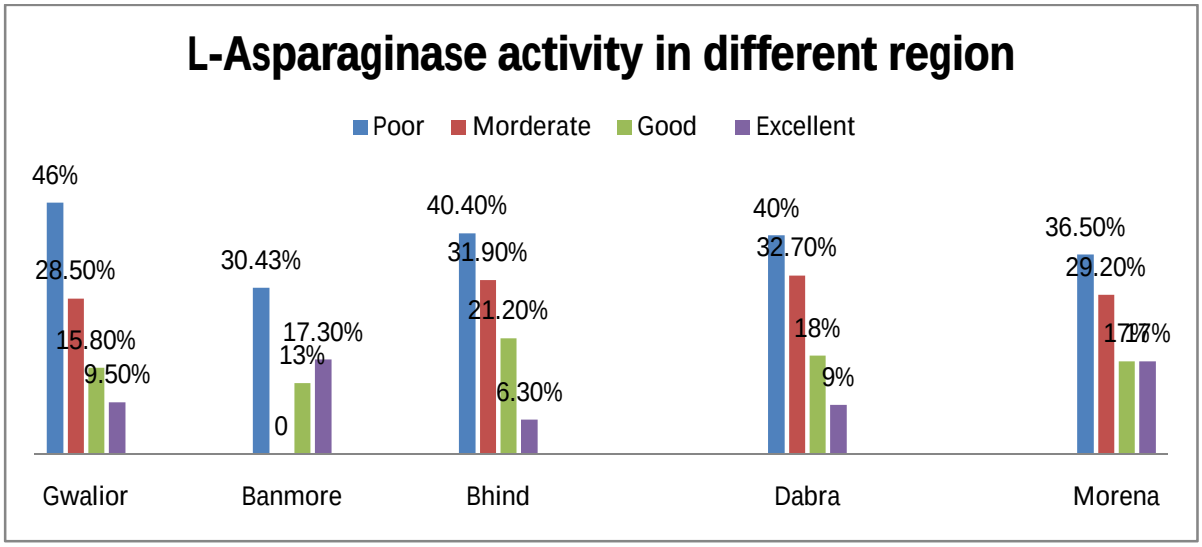


Fig 4 : Quantitative status of L-Asparaginase producing isolates in different region of Madhya Pradesh

IV. DISCUSSION

The findings of the present work revealed that the bacteria isolated from Madhya Pradesh, India have a potentiality for extracellular L-Asparaginasee production that shows futuristic approach for the purification studies and evaluation of its antitumor activity. Present study was targeted for the search of new bacterial species with having demandable enzyme characteristic objective of present work was to isolate and screen the L- Asparaginase producing bacteria from the soil of Madhya Pradesh. For that soil samples of different region of Madhya Pradesh, like Gwalior, Bhind, Morena, Dabra and Datia were collected. From them 312 soil bacteria were isolated and all isolates were subjected to primary screening based on plate assay protocol. Isolates producing L-asparaginase were identified by a pink coloured colony on modified M9 agar medium with phenol red as an indicator for detection

of L-Asparaginase producing colonies **[10]**. The isolates with the pink coloured colony were selected for further studies, however the organism did not produce any pink zone in both control groups - M-9 media incorporated with L-Asparagine but without phenol red and M-9 media incorporated without L-asparagine. This indicates that the formation of pink zone is due only to L-asparaginase production. For the production of enzyme, it is necessary to know the quantity of enzyme L-Asparaginase produced by soil isolates. The second objective of the study was the quantification of L-Asparaginase produced by all 229 isolates, which shown positive L-Asparaginase production after primary screening. Enzyme activity was estimated by the Nesslerization method. The enzyme activity was expressed in international units per mililiter (IU/ml). The liberation of ammonia can be determined by reaction of Nessler's reagent and the production of yellow color was evaluated. The ammonia content was estimated by a standard curve prepared from various dilutions of ammonium chloride solution. After quantitative estimation, 229 isolates were categorize in to four groups which shown that 23% isolates with less than 0.1 IU/ml enzyme activity, considered as poor producers, 36 % isolates shown 0.1-0.5 IU/ml activity, as moderate producers, 29% isolates good enzyme producers have enzyme activity in range of 0.5-1 IU/ml and among them maximum activity was shown by 10% isolates above than 1 IU/ml which were considered as excellent enzyme producers.

V. CONCLUSION

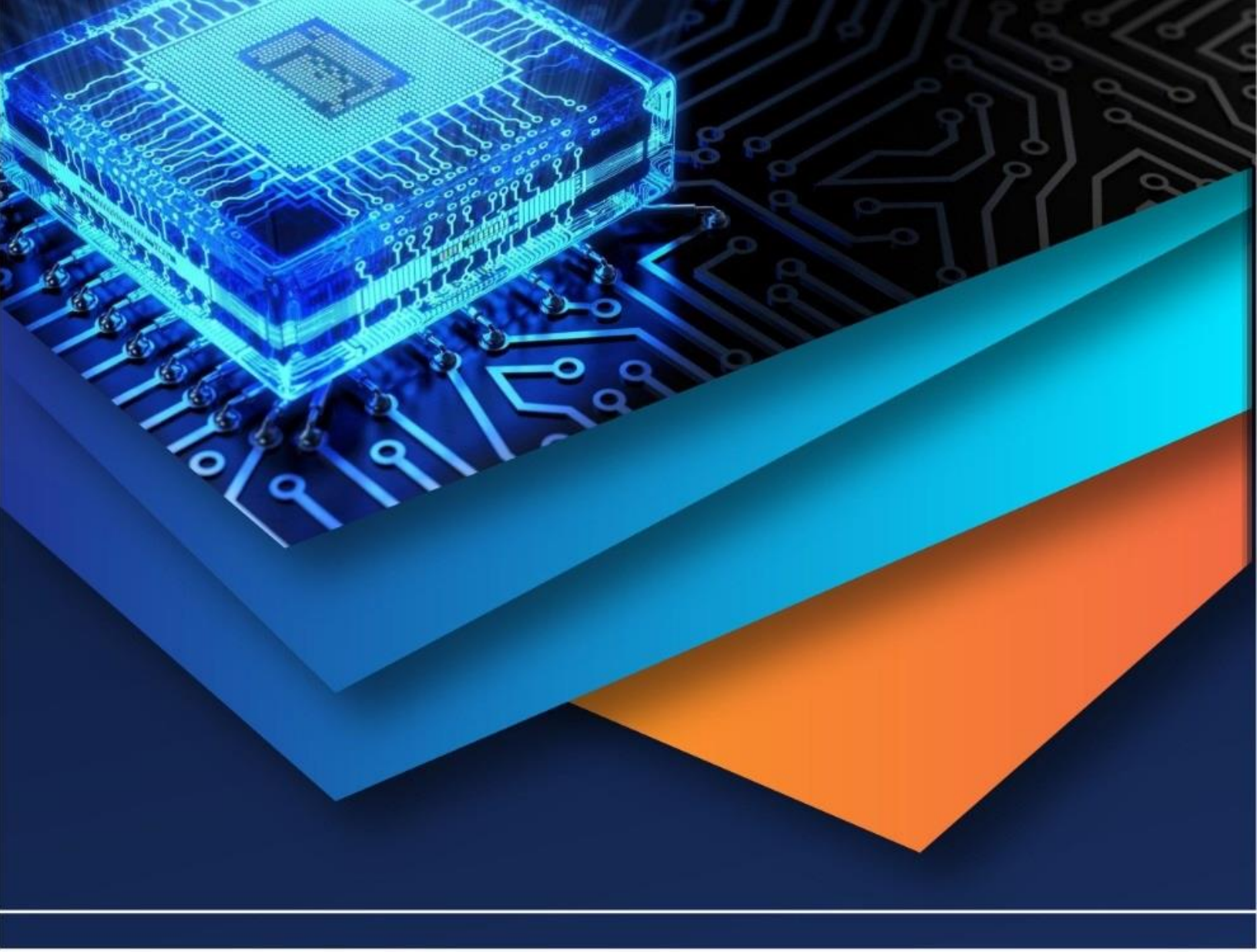
L- Asparaginase is a very important enzyme for commercial purpose used as an anti-neoplastic agent. With the development of its new functions, a great demand for L-asparaginase is expected in coming years Bacteria are promising source of a wide range of important enzymes with respect to their medicinal use, some of which produced in industrial scale. It is necessary for the isolates which gives maximum activity of enzyme should be able survive in different physiological conditions because enzyme produce by this type of isolates may have ability for stable enzyme activity to tolerate different physiologic condition for its industrial applications. There is a immense need of L-Asparaginase which bind substrate efficiently that can be calculate by considerable K_M value of the enzyme, and it can also more efficient in physiological pH. Thus, it is desirable to search for new bacterial isolates producing L-asparaginase with novel properties from as many different sources as possible.

VI. ACKNOWLEGMENTS

Author thankful to a Dr. B.R. Shrivastav, head department of surjury, cancer hospital Gwalior for support during research.

REFERENCES

[1] Pieters, R., Hunger, S.P., Boos, J., Rizzari, C., Silverman, L., Baruchel, A., Goekbuget, N., Schrappe, M. and Pui, C.H., 2011. L-asparaginase treatment in acute lymphoblastic leukemia. Cancer, 117(2), pp.238-249.
[2] El-Bessoumy A A, Sarhan M and Mansour J (2004), "Production, Isolation, and Purification of L-Asparaginase from Pseudomonas Aeruginosa 50071 Using Solid-state Fermentation", J. Biochm. and Mol. Biol., Vol. 37, No. 4, pp. 387-393
[3] Sahu, M.K., Sivakumar, K., Poorani, E., Thangaradjou, T. and Kannan, L., 2007. Studies on L-asparaginase enzyme of actinomycetes isolated from estuarine fishes. Journal of Environmental Biology, 28(2), p.465
[4] Gupta, N., Dash, S.J. and Basak, U.C., 2009. L-asparaginases from fungi of Bhitarkanika mangrove ecosystem. AsPac J. Mol. Biol. Biotech, 17(1), pp.27-30
[5] Nottage, K.A., Ness, K.K., Li, C., Srivastava, D., Robison, L.L. and Hudson, M.M., 2014. Metabolic syndrome and cardiovascular risk among long-term survivors of acute lymphoblastic leukaemia-From the St. Jude Lifetime Cohort. British journal of haematology, 165(3), pp.364-374
[6] MacDonald, R.M., 1986. Extraction of microorganisms from soil. Biological Agriculture and Horticulture, 3(4), pp.361-365
[7] Gulati, R., Saxena, R.K. and Gupta, R., 1997. A rapid plate assay for screening l-asparaginase producing micro-organisms. Letters in applied microbiology, 24(1), pp.23-26
[8] Imada, A., Igarasi, S., Nakahama, K. and Isono, M., 1973. Asparaginase and glutaminase activities of micro-organisms. Microbiology, 76(1), pp.85-99
[9] Lowry, O.H., Rosebrough, N.J., Farr, A.L. and Randall, R.J., 1951. Protein measurement with the Folin phenol reagent. Journal of biological chemistry, 193(1), pp.265-275
[10] Prakasham, R.S., Hymavathi, M., Rao, C.S., Arepalli, S.K., Rao, J.V., Kennady, P.K., Nasaruddin, K., Vijayakumar, J.B. and Sarma, P.N., 2010. Evaluation of antineoplastic activity of extracellular asparaginase produced by isolated Bacillus circulans. Applied biochemistry and biotechnology, 160(1), p.72.



10.22214/IJRASET

45.98

IMPACT FACTOR: 7.129

IMPACT FACTOR: 7.429

INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY