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Isolation and Prelimnary Screening of Microbial Isolates for Extracellular Production of Amylase

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Abstract: The present study evaluated an amylolytic potential of the isolates from selected soil sample. Initial screening process involved a selection medium containing starch as a sole source of collect five soil sample, Flour Mill (FM), Domestic wastes dumped soil (DWDS), Agricultural soil (AS),

Agricultural waste soil (AWS), Prince Bakery waste dumped soil (PBWDS). The soil sampling sites were selected from vicinity of the Bilaspur city naturally rich in starch. The fungus and bacteria isolated were screened on starch agar medium and the zone of hydrolysis was compared for prelimnary selection of the isolates. The molds were found to be the most potent group for Amylase production.

Keywords: Amylase, isolation, prelimnary screening and starch agar medium.

1. Introduction

Sincepast enzymes have been used as an efficient tool in industries for production of economically important product. Amylase is an enzyme that catalyses the hydrolysis of starch into sugars. Enzymes like amylases, carboxymethylcellulases and proteases are widely used in the industry for the manufacture of pharmaceuticals, foods, beverages and confectioneries as well as in textile and leather processing, and waste water treatment [1-2]. The majority of the enzymes used in the industries are of microbial origin because of their more stable nature as compared to the corresponding enzymes derived from plants and animals [3].

2. MATERIALS AND METHODS

- **2.1 Isolation of microorganisms from different soilsamples:** The soil samples for isolation of potent microorganisms were collected from places in and around Bilaspur 1.Flour Mill (FM), 2.Domestic wastes dumped soil (DWDS), 3. Agricultural soil (AS), 4.Agricultural waste soil (AWS),5. Prince Bakery waste dumped soil (PBWDS). The soil sample was collected in sterilized polythene bags.
- **2.2 Isolation and maintenance of the pure cultures:** The isolated colonies were selected on basis oftheir fast growth and isolated on starch agar medium from five selected soil samples. The isolates were stored on slants at 4°C till further use.
- **2.3 Plate assay method for qualitative screening:** The method of Salwa*et al.* (2012) was followed for preliminary screening of the isolates by Starch hydrolysis Test. A blue/black or purple zone surrounding the growth indicates that starch is present and has not been hydrolyzed (-) and the organism did not produce the extracellular enzymes. Colourless zone indicates hydrolysis of starch.

3. RESULTS AND DISCUSSION

The isolation of microbes viz mold, bacteria and yeast were isolated from selected soil sampling sites listed in table.1. The total number of isolates of three microbial categories are given in table 1.

Maximum molds (36) were isolated from DWDS maximum yeasts (20) were isolated from DWDS. Maximum bacteria isolated (50) from agriculture waste soil as given in table 1.

Table -1. Total microbes isolated from selected soil samples

S.No.	Soil Sample	Molds	Yeast	Bacteria
1	Flour Mill (FM)	10	10	15
2	Domestic wastes dumped soil (DWDS)	36	20	11
3	Agricultural soil (AS)	28	12	48
4	Agricultural waste soil (AWS)	15	19	50
5	Prince Bakery waste dumped soil (PBWDS)	12	10	36
	Total	101	71	135

All the microorganism isolated from five selected soil samples were subjected for preliminary screening by plate method. The zone of hydrolysis measured in cm is given in fig.1 –fig.15 for molds, yeast, bacteria.

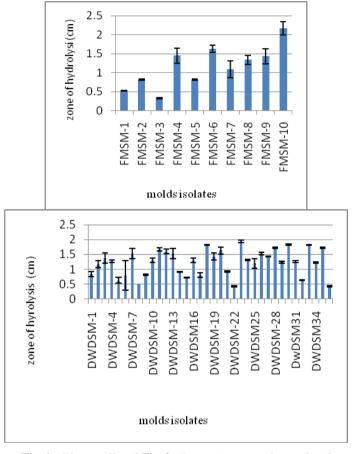


Fig-1. Flour mill soil Fig-2: Domestic waste dumped soil 2 2.5 zone of hyrolysis (cm) zone of hyrolysis (cm) 1.5 1 0.5 AWDSM9 AWDSM13 AWDSM3 AWDSM5 **AWDSM7 AWDSM11** AWDSM15 AWDSM1 ASM13 ASM16 ASM19 ASM28 ASM4 ASM7 ASM10 molds isolates molds isolates

Fig-3. Agricultural soil Fig-4. Agricultural waste dumped soil

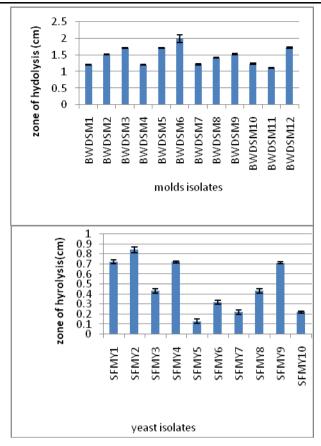


Fig-5. Flour mill soil. Fig-6. Flour mill soil

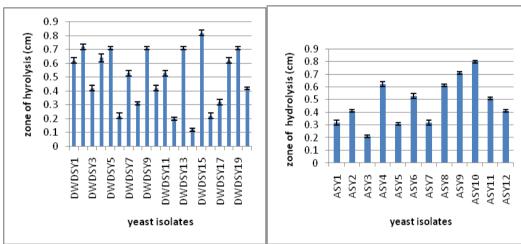


Fig-7. Domestic waste dumped soil

Fig-8. Agriculture soil

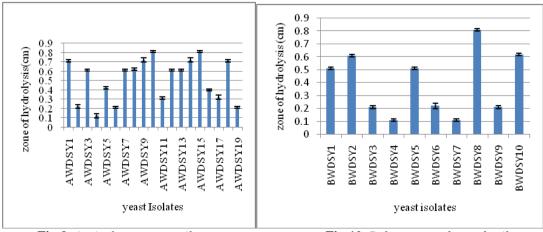


Fig-9. Agriculture waste soil

Fig-10. Bakery waste dumped soil

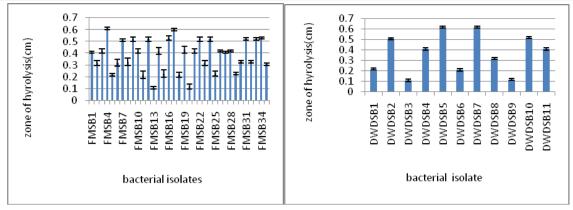


Fig-11. Flour mill soil 0.7 zone of hydrolysis 0.6 0.5 0.4 0.3 0.2 0.1ASB13 ASB29 ASB37 ASB33 ASB21 ASB41 ASB5 ASB17 ASB25 bacterial isolates

Fig-12. Domestic waste dumped soil zone of hydrolysis 0.6 0.5 0.4 0.3 0.2 0.1 AWDSB5 AWDSB29 AWDSB9 AWDSB13 AWDSB17 AWDSB21 AWDSB25 AWDSB33 AWDSB37 AWDSB45 AWDSB49 AWDSB1 AWDSB41

Fig-13. Agriculture soil bacteria

Fig-14. Agriculture waste dumped soil

bacterial isolates

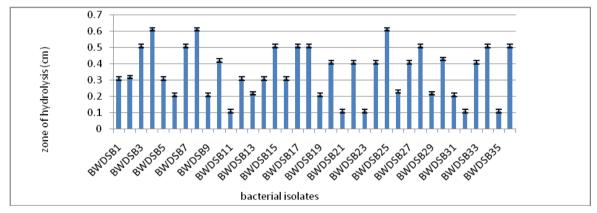


Fig-15. Bakery waste dumped soil

4. DISCUSSION

All the soil samples were excellent sourcesof microbes [4]. The molds were found to be potent Amylaseproducers similar to the earlier findings [5].

The maximum zone of hydrolysis was observed to be 2.17 ± 0.17 cm.comparable to the previous reports [5-6].

5. CONCLUSION

The soil samples proved to be excellent sources of Amylases .The screened organism exhibited a natural potential to produce Amylase which can be increased by optimization and used for industrial applications.

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