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Qualitative Study of Solubilization Activity by Indigenous Isolates

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ABSTRACT

Today as a result of extensive research, the study of microbial products is recognized as an integral component of natural products chemistry and as well as a significant resource for environment friendly compounds. Eco-friendly compounds have multiple industrial applications. This work was carried out to characterize the bacteria from different environmental sources for the production of commercially important products. Various bacteria were isolated from different environmental sources such as soil, fresh water and marine, and also from CMG stock. Among twenty five strains, three bacterial strains, CMG645, CMG646, and DWiwere selected. The selected bacterial strains were studied for solubilization activity which is a commercially important property.

Keywords: *Baitene, Environmental, Metals, Solubilization*

INTRODUCTION

Zinc is an essential micronutrient for microorganisms and plants. It is present on earth's crust in tune of 0.008%. It has an immense role in nutrition of both eukaryotic and prokaryotic organisms as cofactor or metal activator in various enzyme systems (Sharma *et al.*, 2014). The metabolic activity of microorganisms (production of acids) solubilizes phosphate from insoluble calcium, iron, zinc and aluminum phosphates. Phosphates are released from organic compounds such as nucleic acids by microbial degradation (Burgstaller & Shinner, 1993).

Biological processes can solubilize metals, thereby increasing their bioavailability and potential toxicity (White & Gadd, 1997).

Mechanism of solubilization includes few steps, which are interrelated while, solubilization is the result of combine participation of all these mechanism. These methods include proton extrusion, enzymes production, sideropho reproduction, and organic acid production. This paper will focus only on the qualitative screening of solubilization activity by indigenous isolates from Karachi.

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MATERIALS AND METHODS

25 bacterial strains were studied qualitatively for determination of solubilization potential for zinc oxide and zinc phosphate in Tris minimal medium containing glucose as carbon source (2%), Tris 6.06gm, NaCl 4.68gm, KCl 1.49gm, NH_4Cl 1.07gm, Na_2SO_4 0.43gm, $MgCl_2.7H_2O$ 0.20gm, $CaCl_2$ 0.03gm, Na_2HPO_4 40.0gm, NH_4Citrate 02.0gm, distilled water 1L.

Three isolates i.e. DWi, CMG646 and CMG645 were strong bearers of the activity and were used for further study.Solubilization activity was determined by inoculating selected bacterial strains in Tris minimal media in presence of insoluble inorganic metal salts.

At each specific hour along with growth curve

readings, culture of the selected bacterial strains was inoculated in the Tris broth amended with in-soluble inorganic metal salt to check that at which time during the growth the solubilizing compounds appear in the medium andhow long they persist.

RESULTS

Solubilization activity is determined by the appearance of the clear haloes but its intensity is determined by the zone size. On Tris minimal media all the selected bacterial strains showed solubilization zone for their respective metals. Solubilization activity was detected by the disappearance of added mineral particles i.e. insoluble inorganic Zn and production of clear zones around the growth.

Supernatant inoculations during growth showed that solubilization activity does not appear in the supernatant before 4 hrs., after inoculation. In DWi this activity appeared in the supernatant around 8thhr. of the growth and persists up to 30thhr of the growth. In CMG646 it appeared at 8thhr. of growth and persist for 30th hr. of the growth. While in CMG645 it appeared at 16th hr. and persist up to 34th hr. The maximum retention of the solubilization activity in the supernatant is shown by the CMG646 and DWi.

These strains were also tested for production of acids and it was found that these strains were capable of producing acetic acid, while oxalic acid production was not confirmed.

DISCUSSION

Zinc deficiency is major problem that affects soil health and crop yields in agriculture. Microorganisms play an important role in solubilization of zinc in soil. All the bacterial strains were screened for solubilizing insoluble inorganic metal salts. For this purpose bacteria were grown on Tris-minimal medium, in presence of insoluble inorganic metal salts, clear haloes were observed along growth which indicated that bacteria not only capable of growingon Tris medium but also can solubilize the insoluble metal salts. CMG645, CMG646, and DWi were selected for quantitative study of solubilization activity.

Quantitative analysis of solubilization activity showed that in the culture it increases rapidly as the bacteria grow on the plates they solubilize the insoluble, inorganic metal salts, while in case of supernatant the activity was analyzed and detected by inoculating on the plates, supernatant was prepared by filtering from culture after every two hours by Millipore filter paper (0.2µm). In case of CMG646 formation of zone was observed around supernatant spot upto 15th hour of the growth after which it was observed that the zone did not appear, it can be suggested that the strain has used the product itself and the metabolite which was solubilizing the metal salt was finished, so the zones were disappeared.

Such solubilization of zinc compound mediated through production of organic acids and subsequent release of zinc in external environment and bio-accumulation of Zn inside the cells of bacterial species had been reported earlier (Fasim *et al.*, 2002). Our results are in close agreement with the findings of Ramesh *et al.*, (2014).

It is concluded that bacterial isolates CMG645, CMG646, DWi could be exploited for the production of commercially important products.

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