Screening And Evaluation Of Silver Nanoparticles Producing Bacteria From Lonar Lake, India

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Abstract –

Lonar lake is known for unique microbial diversity, which is located at Deccan Plateau of West-centralof India.Water and sediment sample from lake were collected and screened for presence of silver nanoparticle synthesis. All isolates were grown and analyzed on Horikoshi medium B. The antibacterial activity of these crude silver nanoparticles produced by alkaliphilic bacteria were studied against pathogenic bacteria such as Staphylococcus aureus and Escherichia coli.The zone of inhibition shown by isolates code no. W1A, W1B, W1C, and W2B against S. aureus and E. coli was in the range of 20 mm to 28 mm. The zone of inhibition shown by silver nanoparticles is significant in comparison with traditional antibacterial agents.

Introduction –

Lonar Lake (Lonar Crater, Lonar Sarovar) is a notified National Geo-heritage Monument. It is a alkaline lake located at Lonar in Buldhana District, Maharashtra, India. It was created due to a meteor impact during the Pleistocene Epoch (Antony et al., 2010; Kanekar et al., 2012). A British Officer, C J.E Alexander identified it in 1823. Lonar Crater is filled with saline water and the uniqueness of water is its salinity and high alkalinity (Fredriksson et al.,1973). Alkalinity environments show diverse flora of alkaliphilic microbial culture growing at pH 8-10 and some at high salt concentration.

Nanotechnology has become one of the most promising technologies applied in all areas of science. Currently there is growing need to develop an environment friendly nanoparticles synthesis that does not use toxic chemicals in the process of its synthesis. The microbial mediated biological synthesis of metallic nanoparticles has recently been recognized as a promising source for mining nanoparticles. The microbial recovery of precious metals with the formation of their nanoparticles is a green alternative to the conventional method (Gandhi & Khan, 2016).

Materials and methods –

A) Enrichment, Isolation and identification of bacterial isolates –A total three samples,(two water and 1 sediment) were collected from different sites of Lonar lake. Enrichment of the cultures were out on Horikoshi media A, B and C respectively (Horikoshi K, 1999). After enrichment, the well isolated and differentiated colonies were transferred on respective medium slants and cultures were maintained as stocks. Isolated Bacillus species were identified by cultural, morphological, biochemical tests.

B) Synthesis, Characterization and Antibacterial activity of crude silver nanoparticles (AgNPs) – Total 8 isolates collected from Lonar lake were sub-cultured in test tube containing 10 mL of nutrient broth containing 3.5 mM AgNO3. The inoculated broth incubated at dark condition at room temperature for 15 days. After incubation period upon visual observation, the culture incubated in presence of silver nitrate. Along with these the control experiment was also run without AgNO3.The biosynthesized silver nanoparticles from bacteria isolated from Lonar lake were screened against one Gram – positive and one Gram - negative bacteria such as S. aureus and E. coli respectively. The method used for antibacterial potential was well diffusion method on nutrient agar. Zone of inhibition showed by silver nanoparticles against pathogenic bacteria were measured.

Result and Discussion

In the present study, total three samples comprising of two water and one sediment were collected from different sites of alkaline Lonar Lake, India. In the winter season December 2018. From these samples 8 morphologically different colonies were isolated. Out of these 8 isolates 4 isolates produce significant amount of silver nanoparticles. After synthesis of Agnp's supernatant used for the antibacterial activity.

Table 1 - Zone of inhibition against *S.aureus & E. coli*

Sr No.	Culture code	Zone of inhibition showed by Bacterial suspension (mm)		Zone of inhibition showed by Bacterial AgNPs (mm)	
		against S.aureus	against *E. coli*	Against S.aureus	against *E. coli*
1	W1A	No zone	No zone	28 mm	20 mm
2	W1B	No zone	No zone	28 mm	20 mm

3	W1C	No zone	No zone	25 mm	20 mm
4	W2B	No zone	No zone	25 mm	20 mm

Discussion –

Kanekar et al.,(2008) worked on the Lonar Lake, India to identified the bacterial diversity present in the lake. They collected water and sediment sample from the various sites of lake. Isolation of bacteria from samples was performed using Enrichment culture technique. 16S rRNA sequencing and phylogenetic analysis were carried out. Alkalibacillus haloalkaliphilus was the first report of obligately alkaliphilic organism from Lonar lake. In present study 16S rRNA sequencing phylogenetic analysis were also carried out . In the work of Tayde, (2012) Antibacterial Potential of silver nanoparticles produced from Lonar Lake Bacilli, Bacilli collected from Lonar Lake were studied. The isolates were grown on nutrient agar containing 3.5 mM AgNO3 under dark condition. In the present study, four silver nanoparticles synthesizing bacteria were isolated. The bacterial isolated were incubated in the presence of AgNO3 for 15 days after 15 days the colour of broth were changed yellow to brown. The supernatant was used further for antibacterial activity against pathogenic bacteria. Rathod et al.,(2016), the actinobacterium Nocardiopsis valliformis OT 1 strain isolated from soil collected from the rim of Lonar Lake. In present study silver nanoparticles producing bacterium were isolated from water sample of Lonar crater. The four isolates were showed efficient production of AgNPs.

Conclusion –

From above results, it was concluded that the bacteria were present in alkaline Lonar Lake has great potential. These isolates were efficient for production of silver nanoparticles.. Our study provides primary evidence that Lonar Lake isolates were promising source for silver nanoparticles as antimicrobial substance. The biosynthesis of silver nanoparticles from Lonar Lake bacteria is eco- friendly and commercially easy process and it can potentially recovers the severe problems caused by chemical antimicrobial agents.It is need to be further studied for its potency and broad spectrum activity for improvement in the antimicrobial activity and production of new drugs.

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