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Reduction of Heavy Metal Toxicity in Cement Using Microorganisms

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Abstract: *A significant material that is frequently utilised in the construction industry is cement. Research indicates that they are restricted to occupational dermatitis. Among construction workers, it is one of the most frequently reported health issues. This is because cement contains heavy metals like nickel (Ni), copper (Cu), iron (Fe), lead (Pb), and magnesium (Mg). The major goal of this study is to use microbes to reduce the toxicity of heavy metals in cement or concrete. The removal and/or recovery of hazardous metals from cement using current technologies is being challenged by the potential of biosorption. The utilisation of biosorption technology's ability to reduce the concentration of heavy metal ions to extremely low levels is one of its main benefits. Heavy metal concentration levels of heavy metals were determined by using Atomic Absorption Spectrophotometry.*

Keywords: *adsorption, bacteria, biosorption, heavy metal, toxicity*

1. Introduction

A significant amount of waste containing heavy metals is produced regularly by the cement- making sectors. Several heavy metals, some of which are poisonous in nature, are used in the production of cement, which serves as the binding ingredient. Dermal problems in construction workers are impacted by heavy metal poisoning. Because the amount of heavy metals in trash varies greatly and may go above the environmentally acceptable limit, they constitute a serious threat to human health. Heavy metal pollution is one of the most serious environmental problems. Metal, a resource that is becoming increasingly rare and seriously pollutes the environment, endangers both the ecology and human health. Metals like mercury (Hg), chromium (Cr), lead (Pb), and zinc (Zn) are considered hazardous metals.

The primary raw materials for cement, limestone, and clay or marl, naturally contain varying levels of trace elements and potentially dangerous heavy metals. Fuels used for both secondary and main purposes are yet another source of emissions [1]. Due to their affordability and sustainability, microbial biosorbents can successfully repair heavy metal-contaminated environments [2]. Microbes have a number of mechanisms for sequestering metals that are more effective than human biosorption. Metals and metalloids can be recovered from liquids using microbial biosorption, which employs living or dead biomass and all of its component elements.

Portland cement contains a variety of heavy metals in various amounts. For more than 20 years [7], German authorities have focused on the leaching of these heavy metals from cementitious building materials. This review looks at the sources of dangerous heavy metals and discusses the several kinds of bacteria that can bio-sorb heavy metals [3].

Globally, the three main drivers of heavy metal contamination are industrialisation, intensive agriculture, and rapid urbanization [6]. Rapid urbanisation, industrialization, and intensive agriculture are the main contributors to the contamination of heavy metals on a global scale [4]. The ecology, agricultural goods, water quality, soil microbes, and human health may all suffer from heavy metals' persistence. Heavy metal contamination is currently one of the largest environmental issues due to the non-degradable properties of metal ions [5]. Environmental contaminants or pollutants are chemicals that are more prevalent in some environmental regions than others [8,9]. Heavy metals are divided into two groups: those with high atomic weights and those with high densities. Metalloids and metallic chemical components are now dangerous for both humans and the environment [10, 11].

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Some recognised conventional methods for heavy metal removal and/or recovery from solution include adsorption procedures, chemical oxidation or reduction reactions, chemical precipitation, electrochemical methods, evaporative recovery, ion exchange, reverse osmosis, and sludge filtration [12, 13]. Heavy metals may gradually become more poisonous, infiltrate the food chain, and impact people and other animals, demanding their removal from the polluted environment [14]. Heavy metals in the soil can have a number of negative consequences on plants and plant seeds, including impaired soil respiration, physiological dysfunction, and hunger, as well as accumulation in the body and possibly irreversible repercussions on human health [15].

Excavation, solidification, and stabilisation, as well as additional methods such as chemical adsorption, are the classic methods for removing heavy metals from polluted areas [16, 17]. The drawbacks of these methods are their high cost, the creation of dangerous secondary metabolites, and their inefficiency [18]. They also temporarily eliminate heavy metals. Contrarily, biological solutions address these issues because they are easy to use, inexpensive, and environmentally beneficial because they don't generate secondary pollutants. Moreover, they support the preservation of soil structure and almost completely remove pollutants and germs from a polluted environment [19]. The biochemical characteristics, physiological adaptation, and/or genetic makeup of the bacteria, which includes morphological and environmental changes to metal speciation, are all necessary for their survival in heavy metal-polluted soils [20]. It was shown that Aspergillus fumigatus had good biosorption abilities for a number of heavy metals. Microorganisms can actively reduce heavy metals through bio-accumulation or passively (biosorption) [21].

Atomic Absorption Spectrometry (AAS) is used to assess the heavy metal toxicity found in the manufactured Ordinary Portland Cement. Significantly, the toxicity hazards associated with the presence of heavy metals in cement may endanger the construction workers (dermal problems), and resident’s health, particularly those of young children. The objective of this paper is to reduce the heavy metals toxicity in cement or concrete. This research aims at using isolated microbial species are used for removing or reducing heavy metals toxicity present in ordinary portland cement (Grade 53). This research work, some isolated microbial strains have been used for heavy metal toxic removal ability discussed. This investigation ultimately gives the solution for the construction industries and cement manufacturing industries.

2. Materials and methods

2.1. Methodology

The process of the research work is shown in Figure 1.

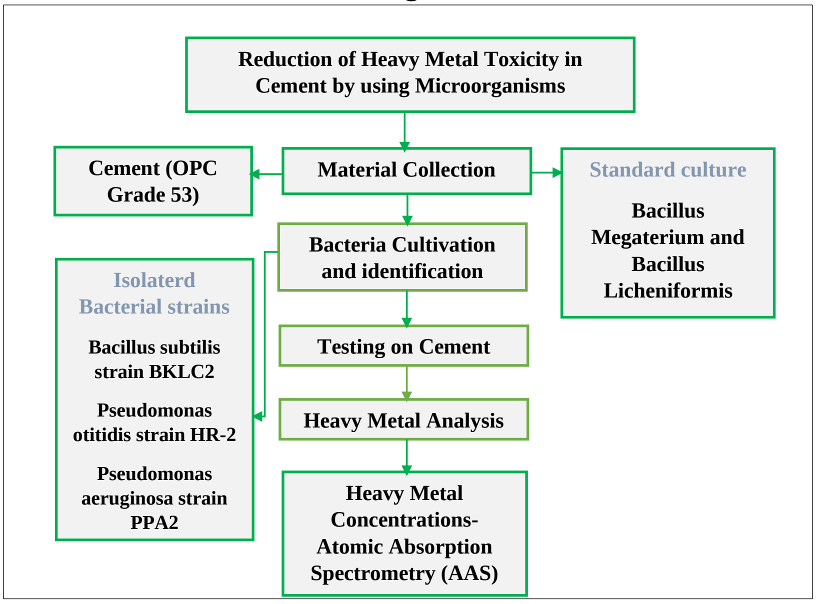

Figure 1. Methodology

2.2. Material collection

For this research Ordinary Portland Cement (Grade 53) *Dalmia Cement* was purchased in Coimbatore, Tamilnadu, India. Standard cultures of *Bacillus Megaterium (NDRI-067)* and *Bacillus licheniformis (NDRI-598)* were bought from the National Dairy Research Institute (NDRI), Karnal, India.

2.3. Bacteria cultivation

Organisms were isolated from the microbial consortium sample. Microorganisms were cultivated by using the serial dilution method. Potential bacteria's identified by using the 16S rRNA gene sequencing technique.

2.4. Tests on cement

2.4.1. Setting time test

The initial setting time of concrete is the period from the addition of water to the cement until a 1 mm square section needle cannot pierce the cement paste that has been placed in a Vicats mould with a bottom thickness of 5 to 7 mm.

2.4.2. Fineness test

Cement fineness, which is expressed in terms of the specific surface area of cement, is a measurement of cement particle size. The cement sample is sieved via a standard IS sieve for the fineness test. Calculating the percentage of retained cement particles requires determining the weight of cement particles with a size greater than 90 microns.

2.4.3. Soundness test

The Soundness of cement can be defined as its ability to retain its volume after it gets hardened. This means that a properly sound cement will undergo minimum volume change after it gets converted in a hardened state. In the soundness test of cement, we determine the amount of excess lime.

2.4.4. Consistency test

The consistency of cement test is carried out to ascertain how much water should be added to cement to achieve standard consistency or normal consistency. Cement begins to hydrate when combined with water. When less water than necessary is added, cement is improperly hydrated and loses strength. Standard or Normal Portland cement has a consistency that ranges from 25 to 35%. The capacity of the substance to withstand failure in the form of fissures and cracks determines the compressive strength.

2.5. Heavy metal analysis

Atomic absorption spectroscopy was used to examine Mg, Fe, Cu, Pb, and Ni (ICE 3000 AA01191606 v1.30). The acid extraction was carried out the method 3050B (USEPA, 1996). For digestion, 1g of the sample was poured into a 250 mL flask. In 10 mL of 50% HNO_3, the material was heated to 950ºC. After getting it to cool, it was refluxed several times with additions of 60% HNO_3 until the sample was free of any odours of brown. The solution was given time to evaporate until 5mL persisted. The mixture was once again incubated with 10mL of 37% HCl for 15 min. The digestate was collected, filtered through a 0.45µm centrifuge tube, diluted to 150 mL with deionized water, and kept at 4°C for evaluation. Atomic absorption spectroscopy was used to measure the heavy metal contents. (AAS machine: ICE 3000 AA01191606 v1.30 Atomic Absorption Spectrophotometer). Table 6 illustrates the Heavy metal concentrations mg/L(ppm). The following formula was used to compute the substantial reduction in metal concentrations.

[(Initial Concentration of metal-final concentration of metal)/Initial concentration]X100

3. Results and discussions

3.1. Properties of cement

OPC Grade 53 was used for various tests such as setting time test, consistency test, soundness test, fineness test and strength test as it is the most used grade in construction. The properties of cement are illustrated in Table 1 and also the compressive strength of the bacterial concrete strength achieved 28 days at 53.48 N/mm^2. As per IS 12269 (1987): 53 Grade Ordinary Portland Cement were used.

Table 1. Properties of cement

S.No	Properties/Strength	Results
1	Grade of Cement	OPC 53
2	Initial Setting Time	35 mins
3	Final Setting Time	480 min
4	Fineness modulus	2.78%
5	Soundness	10mm
6	Consistency	31%
9	Compressive Strength after 28 days	53.48 N/mm^2

3.2. Heavy metal concentrations - AAS analysis

Table 2 shows the various heavy metal concentration (Mg/L) levels in ppm. The concentration levels of heavy metal reduction after 12 weeks Mg, Fe, Cu, Pb. Ni followed by 11.676,24.792,0.145,0.345 and 3.257. The trend is similar to those of Iron, copper, lead and nickel. However, the percentage reduction in the concentration of Copper was much lower than that of Magnesium and Iron. For Mg, Fe, Ni, the three categories were 11.676, 24.792 and 3.257 ppm respectively.

Table 2. Heavy metal concentrations mg/L(ppm)

Samples	Name of the Isolated species	Accession Code	Heavy Metals	PH Contaminations level	Heavy metal concentrations mg/L(ppm) Reduction after 12 weeks
OPC 53	*Bacillus megaterium*	*067-NDRI*	Magnesium	8.2	11.676
	Bacillus licheniformis	*598-NDRI*	Iron	8.3	24.792
	Bacillus subtilis strain BKLC2	*MG914065.1*	Copper	8.2	0.145
	Pseudomonas otitidis strain HR-2	*MT645611.1*	Lead	8.7	0.342
	Pseudomonas aeruginosa strain PPA2	*MT734698.1*	Nickel	8.4	3.257
BCS	*Bacillus megaterium*	*067-NDRI*	Magnesium	8.2	11.257
	Bacillus licheniformis	*598-NDRI*	Iron	8.3	23.506
	Bacillus subtilis strain BKLC2	*MG914065.1*	Copper	8.2	0.309
	Pseudomonas otitidis strain HR-2	*MT645611.1*	Lead	8.7	0.251
	Pseudomonas aeruginosa strain PPA2	*MT734698.1*	Nickel	8.4	0.169

*OPC – Ordinary Portland Cement, BCS -Bacterial Cement Sample
NDRI- National Dairy Research Institute (Standard culture)

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Table 3. Reduction after 12 weeks

S.No	Name of the Isolated species	Accession Code	Heavy Metals	Reduction after 12 weeks (%)
1	*Bacillus megaterium*	067-NDRI	Magnesium	42
2	*Bacillus licheniformis*	598-NDRI0	Iron	20
3	*Bacillus subtilis strain BKLC2*	MG914065.1	Copper	10
4	*Pseudomonas otitidis strain HR-2*	MT645611.1	Lead	13
5	*Pseudomonas aeruginosa strain PPA2*	MT734698.1	Nickel	15

Figure 2 illustrates the metal reduction after 12 weeks (%) using the Isolated bacterial strains such as *Bacillus subtilis 10%* (Cu), *Pseudomonas otitidis 13 % 15%* (Ni) and standard cultures of using *Bacillus megaterium* (Mg) 42% and *Bacillus licheniformis* (Fe) 20% respectively.

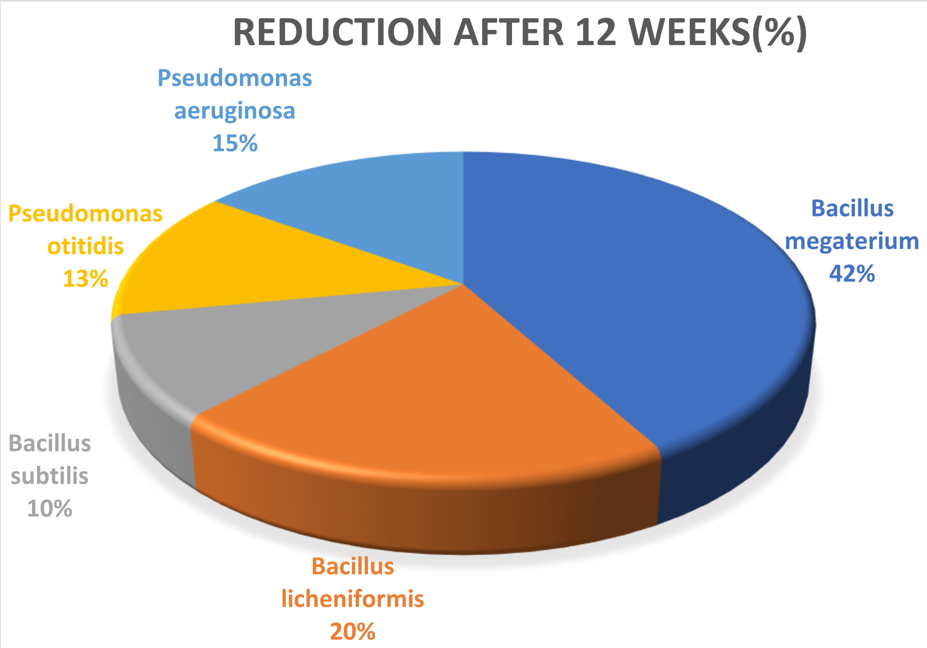

Figure 2. Heavy metal Concentrations-Reduction after 12 weeks

The heavy metal concentration and sources of heavy metal were identified by using AAS analysis solution result as shown in Figure 3.

Tables 2 and 3 show the Copper, lead, and Nickel concentrations of the OPC were 0.145,0.342, and 3.251 ppm respectively. The standard sample OPC-01 had the following values (11.676, 24.792, 0.145, 0.342, 3.57). Iron is found in the highest concentrations with a value of 24.792 ppm compared to existing samples. The highest concentration level in Mg was identified as 11.676 ppm, while the lowest level is identified as 0.51 ppm in BCS. The concentration of lead was 0.342 ppm which is a major toxic metal present in the conventional sample and compared with the bacterial cement sample identified as 0.251 ppm. The reduction levels of metals significant difference between standard cultures and isolated microbial strains. Figure 2 represents the heavy metal toxicity after 12 weeks, pure cultures of Bacillus megaterium(067-NDRI)(Mg) and Bacillus licheniformis(598-NDRI)(Fe) reduced by 42%, 20%. Isolated bacterial strains such as *Bacillus subtilis strain BKLC2(*MG914065.1)(Cu), *Pseudomonas otitidis strain*

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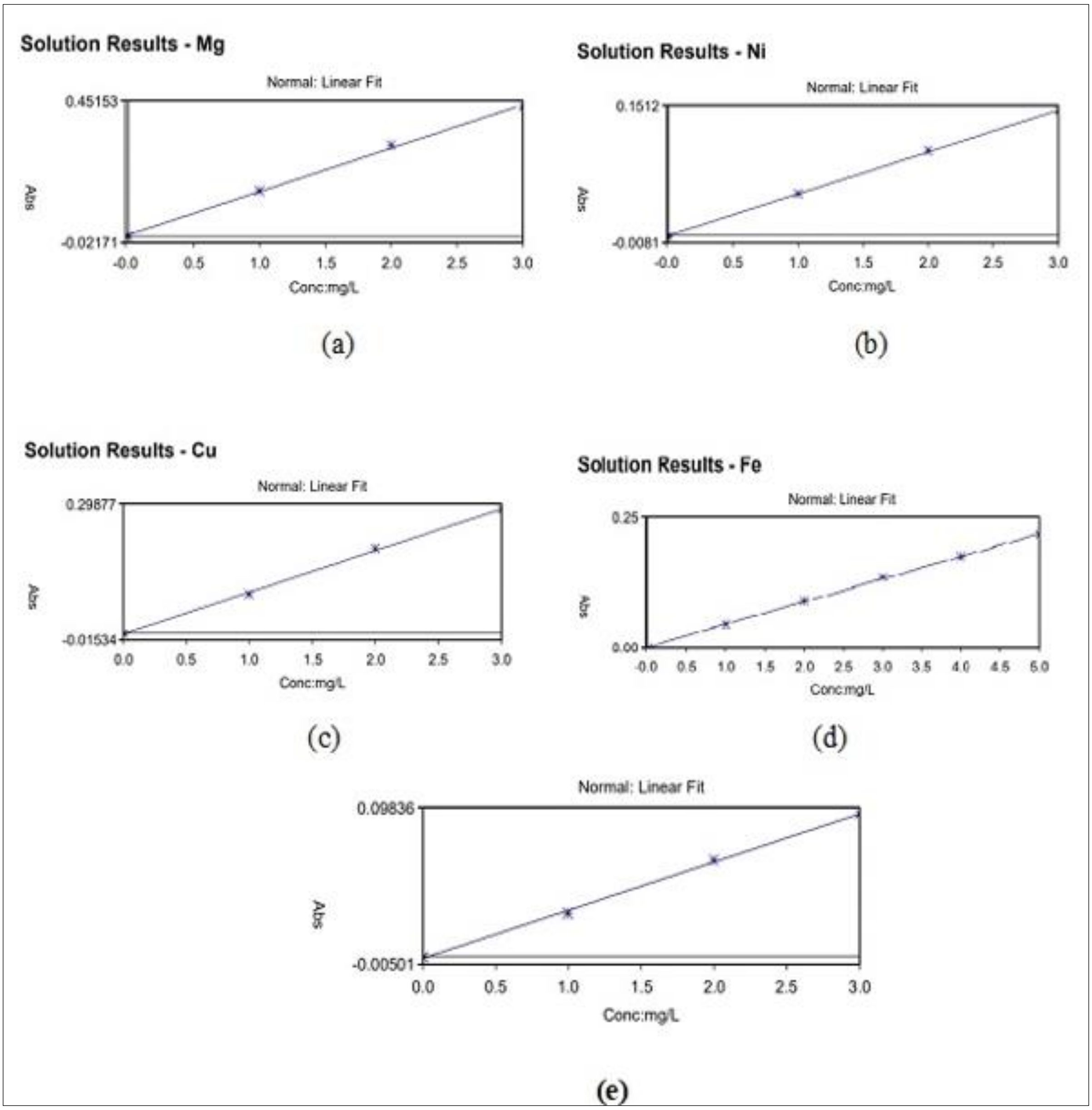

Figure 3. Heavy metal concentrations

*HR-2(*MT645611.1)(Pb) and *Pseudomonas aeruginosa strain PPA2(*MT734698.1) (Ni) toxicity level reduced by 10,13 and 15% respectively. Reduction of toxic heavy metal value compared with isolated strains standard culture produced good results. The strength of our research work mechanical properties of the bacterial concrete strength achieved 28 days at 53.48 N/mm^2, major role of the microorganisms produced good results in concrete and also reduce the heavy metal toxicity in cement. The limitation of the work cultivation and the growth of the microbes are not suitable for all climatic conditions.

4. Conclusions

All the organisms were able to reduce the heavy metal toxicity in cement. The potential micro-organisms were identified using the 16S rRNA gene sequencing technique. Heavy Metal Toxicity analyses were performed on the various concentration levels of heavy metals Magnesium (Mg), Iron (Fe), Copper (Cu), Lead (Pb), and Nickel (Ni) to reduce the toxicity using the isolated bacterial strains and standard cultures. The microbial consortia were able to reduce the heavy metal toxicity of the cement. From the analysis, there is a major reduction of the heavy metal using *Bacillus megaterium (Mg) 42% and Bacillus licheniformis(Fe) 20%* reduced. Isolated bacterial strains such as *Bacillus subtilis* *strain BKLC2(*MG914065.1)(Cu), *Pseudomonas otitidis strain HR-2(*MT645611.1)(Pb) and *Pseudomonas aeruginosa strain PPA2(*MT734698.1) (Ni) toxicity level reduced by 10,13 and 15% respectively. Hence it is concluded that bacterium belonging to the bacillus family is suitable for the reduction of heavy metals toxicity in cement. In cement industries, mainly construction workers face dermal problems and environmental pollution. These microbial consortia isolated species are suitable for reducing the toxic effect of lead by 13%. In future studies identify the remaining toxic heavy metal, remove it in cement and use existing microorganisms.

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