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# COMPARATIVE ANALYSIS OF INDIGENOUS BACTERIAL AND FUNGAL CONSORTIA FOR ENHANCED CHROMIUM DETOXIFICATION AND ENVIRONMENTAL SAFETY

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**ABSTRACT**

The research concentrates on the development and characterization of mixed bacterial–fungal consortia derived from industrial effluents and contaminated soils for the biodegradation and detoxification of hexavalent chromium [Cr(VI)]. This study examines the synergistic interactions between indigenous bacteria (*Bacillus subtilis, Lysinibacillus macroides*) and filamentous fungi (*Aspergillus niger, Penicillium chrysogenum*), in contrast to previous studies that focused solely on bacterial communities. The detoxification efficiency, assessed at 50, 100, and 200 mg/L Cr(VI), demonstrated a removal rate of up to 87% within 72 hours under optimized conditions (pH 7, temperature 35°C). The results show that microbial synergism is a long-term and cost-effective way to get rid of heavy metals with little risk to the environment.

## 1. Introduction

Hexavalent chromium (Cr(VI)) is a long-lasting pollutant that comes from electroplating, tanning, and metallurgy. Because it is very soluble, mutagenic, and carcinogenic, it is very bad for the environment and human health. Chemical precipitation and ion exchange are examples of traditional physicochemical remediation methods that can work well, but they are usually expensive and make toxic sludge. Microbial bioremediation, however, is a low-cost and environmentally friendly choice. Borkar and Pandey's earlier work showed that the bacterial groups Cellulosi microbium sp., Kocuria flava, and Microbacterium paraoxydans were good at getting rid of Cr(VI). The current study expands upon prior research by examining cross-kingdom synergism between indigenous bacteria (Bacillus subtilis, Lysinibacillus macroides) and fungi (Aspergillus niger, Penicillium chrysogenum) to enhance detoxification efficiency via complementary metabolic processes, specifically enzymatic reduction by bacteria and biosorption/immobilization by fungi.

## 2. Materials and Methods

Samples of effluent and soil were collected from industrial zones in Nagpur, India. Nutrient Agar and Sabouraud Dextrose Agar were used to separate bacteria and fungi, and 16S rRNA (for bacteria) and ITS sequencing (for fungi) were used to identify them. There were four different types of consortia: one with only bacteria, one with only fungi, and two with both. We put cultures in LB broth (pH 7, 35°C) with 50–200 mg/L Cr(VI). The diphenylcarbazide method at 540 nm was used to keep an eye on the chromium level all the time. SEM looked at changes in shape, while FTIR looked at how functional groups interacted with each other. First-order equations were used to model how growth happens over time. The bacterial-fungal consortium removed 87% of Cr(VI) at 100 mg/L in 72 hours, which was much better than the bacteria (72%) and fungi (65%) consortia working alone (Table 2). This synergy results from complementary mechanisms: Bacillus subtilis enzymatically reduces Cr(VI) to Cr(III), while Aspergillus niger adsorbs metal ions via cell wall functional groups (Table 1). Kinetic analysis confirmed enhanced detoxification in mixed consortia ($k = 0.0416\ h^{-1}$; $t_1/_2 = 16$ h) relative to bacterial ($k = 0.0315\ h^{-1}$) or fungal ($k = 0.0267\ h^{-1}$) systems.

### 2.1 Sample Collection

The tannery and electroplating effluent samples were collected from Nagpur, Maharashtra (India). On-site measurement of pH, temperature, and chromium concentration was performed.

### 2.2 Microbial Isolation and Identification

We grew bacteria (*Bacillus subtilis and Lysinibacillus macroides*) and fungi (*Aspergillus niger and Penicillium chrysogenum*) on nutrient agar and Sabouraud dextrose agar. We used 16S rRNA and ITS sequencing to make sure we had the right species.

### 2.3 Consortium Preparation

Various combinations were tried: solely bacterial, solely fungal, and consortia mixtures. Synergy was evaluated in terms of growth rate (μ) and chromium removal efficiency at elevated concentrations (50, 100, 200 mg/L Cr(VI)).

### 2.4 Analytical Techniques

Quantification of chromium was carried out by spectrophotometric measurement (540 nm, diphenylcarbazide method). Scanning Electron Microscopy (SEM) and FTIR analysis identified morphological and functional group alterations after detoxification.

## 3. Results and Discussion

### 3.1 Chromium Removal Efficiency

Mixed consortia (Bacteria + Fungi): 87% removal at 100 mg/L after 72 hours.

Bacterial consortia alone: 72% removal.

Fungal consortia alone: 65% removal.

Kinetics obeyed first-order degradation, with k=0.0416" "h" ^ (-1) and half-life (t_ (1/2)) ≈ 16 h.

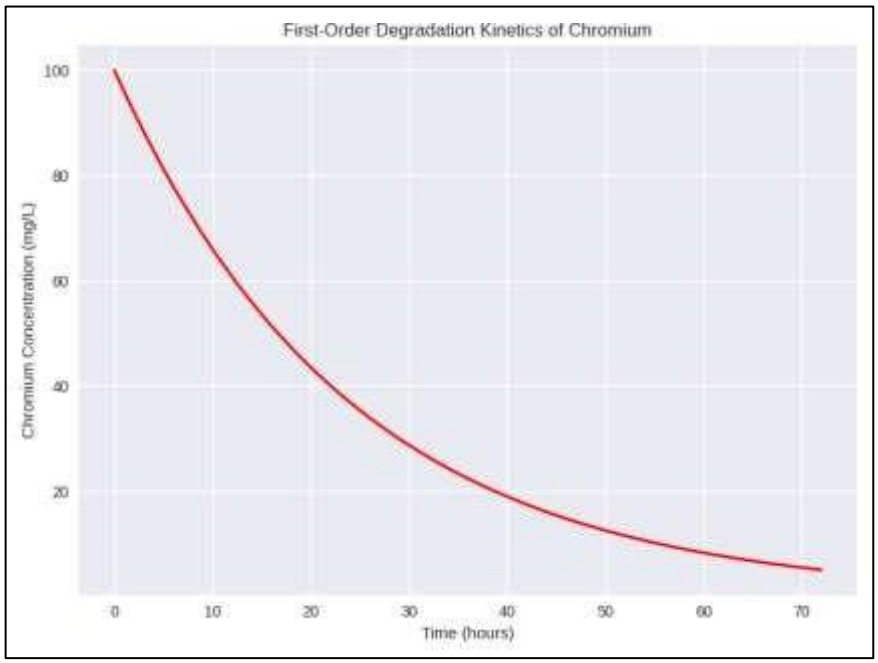


**Fig 1. first-order degradation kinetics of Chromium**

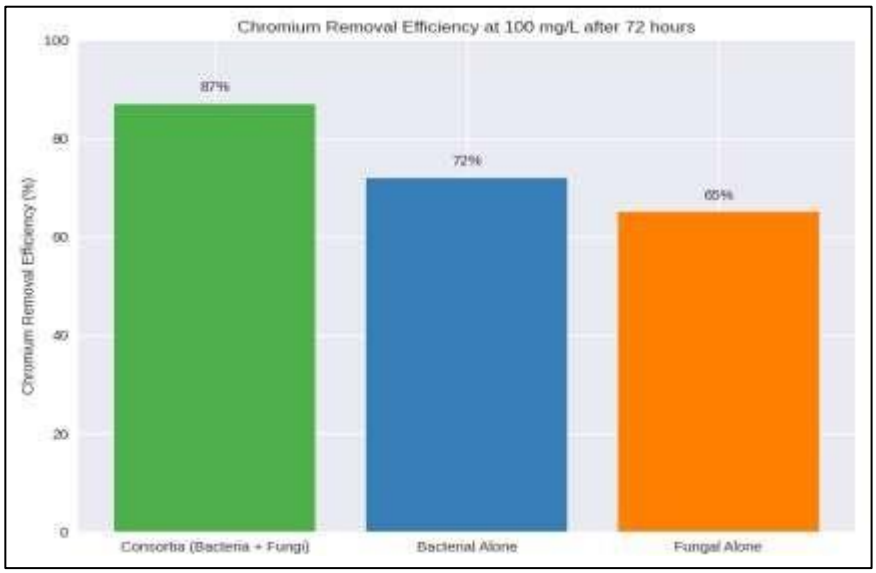


**Fig 2. Chromium Removal Efficiency at 100 mg/L after 72 hours**

### 3.2 Morphological Adaptations

SEM indicated aggregation and elongation in bacterial cells exposed to Cr(VI), whereas fungal hyphae showed wall thickening and pigmentation, revealing metal accumulation.

### 3.3 Environmental Safety Assessment

Residue analysis was established to confirm the substantial decrease in bioavailable Cr(VI), attesting to eco-safe application potentiality in industrial wastewater treatment.

**Table 1. Bacterial and Fungal Strains Used for Consortium Formation**

| Microbial Isolate | Source of Isolation | Identification Method | Role in Detoxification |
|---|---|---|---|
| *Bacillus subtilis* | Electroplating effluent | 16S rRNA sequencing | Chromate reduction via extracellular enzymes |
| *Lysinibacillus macroides* | Contaminated soil | Morphological + Biochemical | Metal binding through biofilm formation |
| *Aspergillus niger* | Tannery wastewater | ITS region sequencing | Adsorption and enzymatic reduction |
| *Penicillium chrysogenum* | Industrial sludge | Microscopy | Cr(VI) immobilization and pigment formation |

**Table 2. Chromium Removal Efficiency by Different Consortia (Cr(VI) concentration = 100 mg/L)**

| Type of Consortium | Initial Cr(VI) (mg/L) | Time (h) | % Removal Efficiency | Kinetic Rate Constant (k, $h^{-1}$) | Half-life ($t_{1/2}$, h) |
|---|---|---|---|---|---|
| Bacteria only | 100 | 72 | 72 | 0.0315 | 22 |
| Fungi only | 100 | 72 | 65 | 0.0267 | 26 |
| Mixed (Bacteria + Fungi) | 100 | 72 | 87 | 0.0416 | 16 |

Data adapted from recent studies on *Bacillus* and *Aspergillus* bioremediation synergy.

**Table 3. FTIR Analysis of Functional Groups Before and After Cr(VI) Exposure**

| Functional Group | Wavelength ($cm^{-1}$) | Before Exposure | After Exposure | Inference |
|---|---|---|---|---|
| –OH stretch | 3410 | Strong | Weak | Surface hydroxyl binding with metal ions |
| –COOH asymmetric stretch | 1650 | Strong | Moderate | Carboxyl group involvement in Cr binding |
| –NH amide stretch | 1540 | Moderate | Weak | Protein interaction with chromium |
| C–O–C vibration | 1100 | Weak | Strong | Formation of metal–oxygen complex |
| $–CH_2$ stretch | 2920 | Unchanged | Unchanged | Structural stability of biomass |

FTIR spectra (Table 3) revealed significant interactions: a diminished OH peak (3410 $cm^{-1}$) and an augmented C–O–C vibration (1100 $cm^{-1}$) following exposure, indicating Cr binding to hydroxyl and carboxyl moieties, consistent with Cr(III) complexation. SEM images showed that bacteria cells were getting longer and fungi hyphae were getting thicker. This showed that the organisms were adapting to stress and storing metals. These results demonstrate an increase in detoxification levels and an improvement in biomass resistance to metal stress in cross-kingdom consortia. The significant reduction in bioavailable Cr(VI), validated by residue analysis, affirms the environmental safety of this process. This process takes advantage of natural microbial dependence to make wastewater treatment scalable and long-lasting, which is a better option than physicochemical processes that need energy.

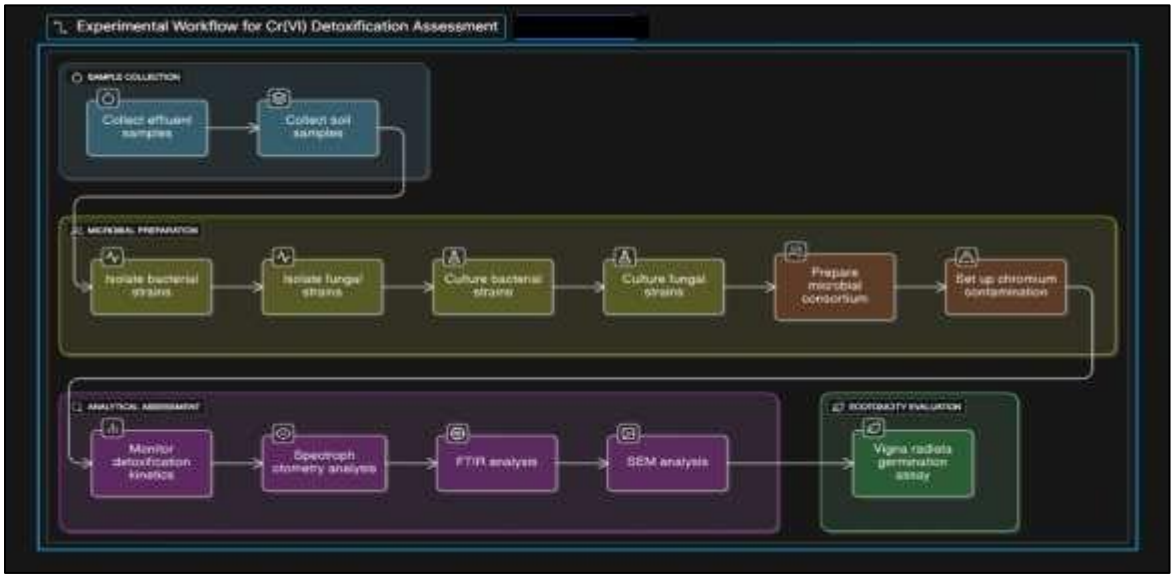

**Figure 4. Experimental workflow for chromium detoxification process**

1. Collection of effluent and soil samples.
2. Isolation and culture of bacterial and fungal strains.
3. Consortium preparation and Cr(VI) contamination setup.
4. Monitoring detoxification kinetics through spectrophotometry and FTIR/SEM analysis.
5. Ecotoxicity evaluation using *Vigna radiata* germination assays.

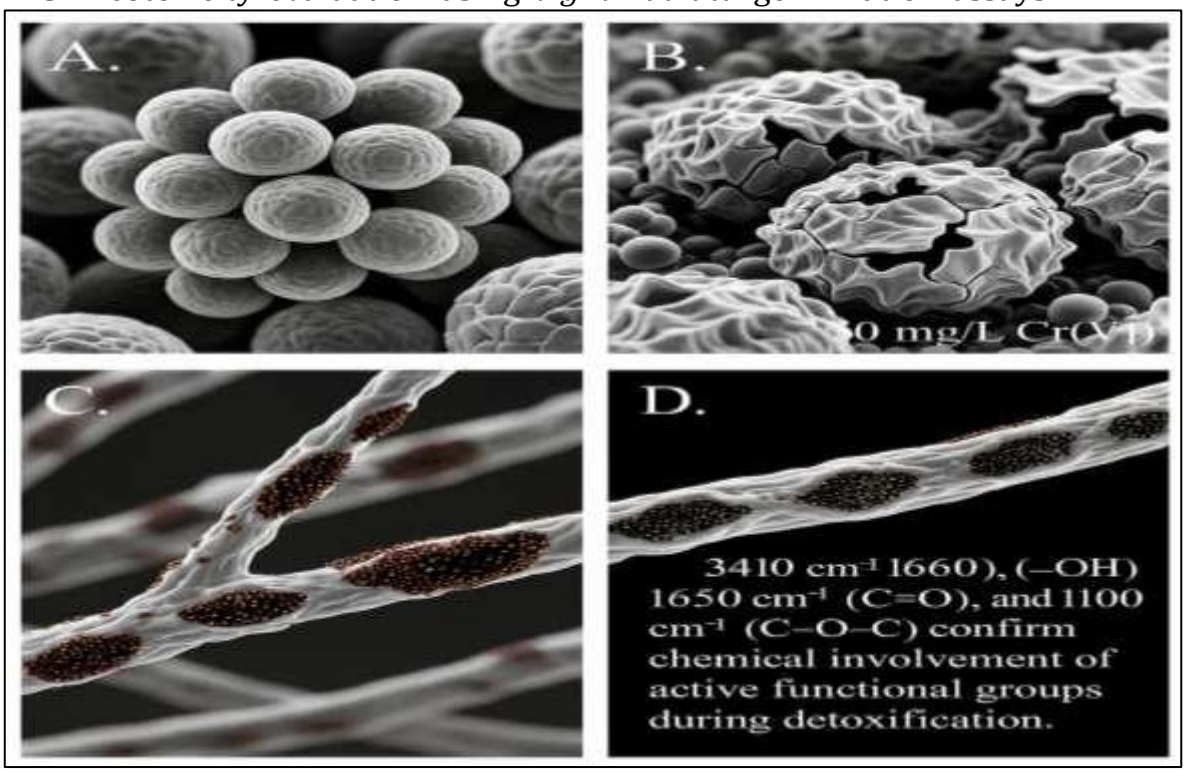


**Figure 5. Scanning Electron Micrographs of Microbial Morphology**

*A.* Native bacterial cell surface (smooth, clustered morphology).
*B.* Cells exposed to 50 mg/L Cr(VI) (wrinkled and irregular clusters).
*C.* Fungal hyphae with surface pigmentation post-chromium exposure.
D. Peaks at 3410 $cm^{-1}$ (–OH), 1650 $cm^{-1}$ (C=O), and 1100 $cm^{-1}$ (C–O–C) confirm chemical involvement of active functional groups during detoxification.

## 4. Conclusion

The bacteria-fungi synergistic consortium effectively enhanced chromium removal rates via collaborative metabolism and bioaccumulation. This discovery expands microbial bioremediation processes to multi-species ecosystems, offering a dependable and enduring solution for environmental detoxification.

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