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COMPARATIVE STUDY ON BIOREMEDIATION OF CHROMIUM IN FORTIFIED SOLUTIONS BY VIABLE, BIOSORBENT AND IMMOBILISED CELLS OF *Phanerochaete chrysosporium*- MTCC787

SUMIT PAL* AND VIMALA Y.

Department of Microbiology, GITAM Institute of Science, GITAM University, Visakhapatnam-530003, AP, India.
*Corresponding Author: Email- sumitmicrobe@gmail.com

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Abstract- Chromium as a toxic heavy metal is of major concern due to the various health effects on human and as a pollutant to environment. Chromium makes land desolate by forming toxic soluble chromium compounds in soil and water bodies. At present various microorganisms are used for bioremediation of chromium from soil and water bodies. The aim of present work was to bioremediate chromium from synthetic solutions by a white rot fungus *Phanerochaete chrysosporium* (MTCC787) using in form of viable cells, microbial biosorbent (dried cells) and immobilised cells at different physical parameters i.e. pH, temperature and medium with substrates like Citric acid, Tween 80 and EDTA.
Key words- Chromium, *Phanerochaete chrysosporium,* Biosorption, Viable cells, Immobilised cells, Tween 80.

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Introduction

Heavy metal contamination is one of the most significant environmental issues, since metals are highly toxic to biota, as they have tendency to accumulate in environment and decrease metabolic activity by unbalancing the ratio of microbial biomass over organic carbon and metabolic quotient 1] and they affect the qualitative and quantitative structure of microbial communities [2-5]. In recent years, the chromium heavy metal pollution becomes a serious and hazardous issue for the human being and biome. The widespread use of chromium and its compounds by modern industries has led to large quantities of this element being released into environments [6,7]. Chromium exists in a variety of oxidation states from 0 to 16 but only hexavalent chromium, Cr (VI), and trivalent chromium, Cr (III), are major concern due to hazardous effect on lithosphere and hydrosphere [8]. It is mutagenic, carcinogenic, and teratogenic, Cr (VI) is about 100-fold more toxic than the trivalent form [9,10]. In United States and Germany, the workers of Chromium -producing industries suffered with Lung Cancer due to exposure of Chromium. [10,11] Cr (III), on the other hand, is considered to be relatively innocuous and even essential to human health in minute quantities[12,13]. Among the different heavy metals, chromium have received special attention due to their strength and persistence in accumulating in ecosystems, where it can cause damage by moving up the food chain to finally accrue in human beings. (Fig. 1) [14-16].

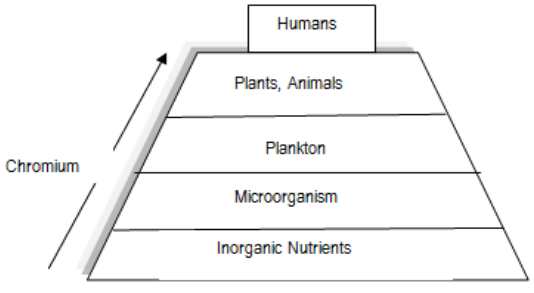

Fig. 1- Accumulation of Chromium through the food chain

Chromium discharged in industrial processes such as wood preserving, metal finishing, Petroleum refining, leather tanning and finishing, paint and ink formulation, manufacturing of automobile parts, mining and surface finishing industry, metallurgy, iron and steel, electroplating, electrolysis, electro-osmosis, photography and electric appliance manufacturing [17-20]. The wastes of these industries containing chromium released directly and indirectly in to environment, having brought serious environmental pollution, and threat to biolife. Several fungi and bacteria has been used for the remediation of chromium like *Aspegillus awamori, Aspergillus flavus, Trichoderma viridae, Pseudomonas* sps*., Pleurotus* sps*, marine algae (e. g. Sargassum natans), Bacillus subtillis, Rhizopus arrhizus and S. cerevisae* [21-24]. These microorganism used as a bioremedient in form of living cells, dry cells and immobilized cells and as a bioremedent they are safe alternative for the physicochemical methodologies. Earlier, White rot fungi i.e. *Pleurotus* have been used to degrade xenobiotic pollutants due its ability to degrade lignin, a polymer in plants. [25,26]. In the same way *Phanerochaete chrysosporium* have manganese peroxidase, lignin peroxidase and laccase enzymes which can helpful in degrading structural polymer in plant and because of this they helpful in degradation of xenobiotic pollutants [27] and various hazardous aromatic compounds in industrial effluents [28,29]. In present study, a comparative study on white rot fungus (*Phanerochaete Chrysosporium*) used for the removal of Chromium from the fortified solutions at different physical parameters i.e. pH, temperature and medium with the different substrate i.e. citric acid, Tween 80 and EDTA to check the enhancement of bioremediation process of chromium under laboratory conditions with three methodologies i.e. viable cells, microbial biosorbent and immobilized cells.

Materials and Methods

Phanerochaete chrysosporium (MTCC787) was collected from MTCC, IMTECH, Chandigarh, India in Lyophilized form and rehydrated by using a Pasteur pipette to add 1 ml sterile water to the freeze-dried pellet then drawn up the entire contents into the pipette and transferred to a test tube with 5 ml sterile distilled water. The fungus was rehydrated for a period of 2 hrs. Then transferred to sterilized malt extract broth and malt extract agar and incubated at the 40°C temperature as given by the MTCC Chandigarh, India. All glassware used for experimental purposes was washed in 10% nitric acid to remove any possible interference by other metals and autoclaved. The media prepared was autoclaved. After 72 hrs of incubation growth appeared, culture was microscopically observed for purity by staining with methylene blue dye.

Preparation of Reagents

1.0 gm of 1-5 Di phenyl carbazide was added in 200 ml of acetone and stored in dried brown colored bottle [30]. Freshly prepared solution was used.

Preparation of chromium solution

282.4 mg of Potassium dichromate was added in 100 ml of distilled water and sterilized at standard condition [30]. The concentration of the Cr (VI) was 1000 ppm in the stock solution. For the test solution, 0.1 ml of the stock solution was added in the 0.9 ml of the medium broth then the final concentration of Cr (VI) was become 100 ppm in the test tube.

Preparation of substrates used as bioremediation enhancer

EDTA Solution- For the preparation of 1M EDTA solution, 21.04 g of EDTA was added in 78.6 ml of distilled water.

Tween 80 Solution- For the preparation of 1% Tween 80 solution, 10 ml of Tween 80 was added in 90ml of distilled water.

Citric Acid Solution- For the preparation of 0.2M of Citric acid solution, 7.3 g of Citrate was added in 92.7 ml of distilled water.

Uptake of Chromium by White Rot Fungus from Synthetic Solutions

Fungal Biosorbent

The culture was grown at 40°C harvested and dried in the hot air oven for 24 hrs at 60^0C [31]. The dried culture weighing 0.2 gms and placed with the test broth i.e. basal salt and malt extract broth prepared by distilled water, 1M Citric acid, 0.2M EDTA and 1% Tween-80 separately at different pH containing chromium solution with control in separate test tubes and incubated at 37^0C and 40° C for 72hrs and centrifuged at 5000 rpm for 10 min and then supernatant taken for the analysis by Di phenyl carbazide method for chromium at 540 nm.

Fungal Viable Cells

In this process the loop full culture of *Phanerochaete chrysosporium* was taken in the test tube containing the basal salt liquid medium prepared with distilled water, 1M citric acid, 0.2M EDTA and 1% Tween-80 separately with dichromate solution in it at different pH 2, 4, 7, 9, 12 and another test tubes as a control. Then these test tubes incubated at room temperature and at 40°C for 72hrs. After 72 hrs the test tubes centrifuged at 5000 rpm for 10 min. and supernatant taken for the analysis by Di phenyl carbazide method for chromium and absorbance was taken at 540 nm in UV - Visible spectrophotometer (Hitachi U-2900).

Fungal Immobilised Cells

In this process the slurry of culture prepared with sodium alginate and poured drop by drop in a 0.1M calcium chloride solution. After formation of beads, it poured in test tubes of different pH containing broths with substrates and dichromate solution and placed at room temperature at 40°C and control in the separate test tubes. After 72 hrs, Chromium absorbance checked at 540 nm by Di phenyl carbazide method.

All the experiments were conducted in triplicates and verified twice.

Calculations

The percentage removal of chromium from fortified solutions was calculated. Percentage removal of chromium is equal to 100 - (A/B×100) where A is optical density of test solution (containing fungus) and B is optical density of control solution.

Results and Discussion

Bioremediation of chromium (VI) was studied using biomass obtained from *Phanerochaete chrysosporium.*

Observations of Bioremediation at Room Temperature and 40°C with Viable Cells at Different pH

In Basal Salt Broth- At pH 2, Chromium was removed 11.02% with distilled water, 10.52% with citric acid, 35.2% with EDTA and

61.86% with Tween 80 at room temperature (Fig. 2a). While at 40°C, with distilled water 46.1%, 28.8% with citric acid, 42.3% with EDTA and 57.33% with Tween 80 (Fig. 2a). At pH 4, Chromium was 38.3% removed in distilled water, 13.88% with citric acid, 11.59% with EDTA and 40.82% with Tween 80 at room temperature (Fig. 2b). On the other hand, at 40°C, 30.9% with distilled water, 28% with citric acid, 32.2% with EDTA and 42.3% with Tween 80 (Fig. 2b). At pH 7, Chromium was 29.2% removed with distilled water, 68.08% with citric acid, 16.43% with EDTA and 41.98% with Tween 80 at room temperature (Fig. 2c).

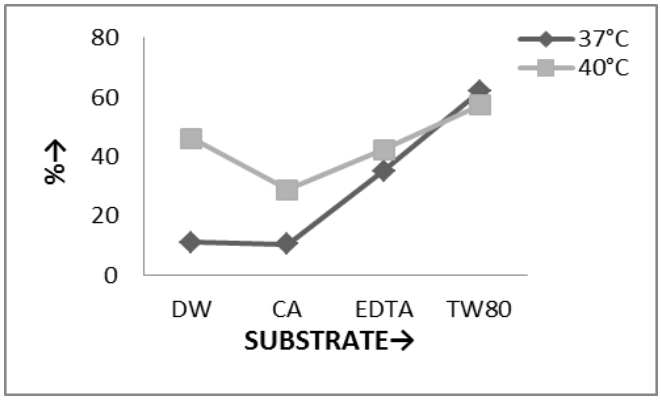

Fig. 2a- Removal of Chromium in Basal salt broth at pH 2 by Viable cells.

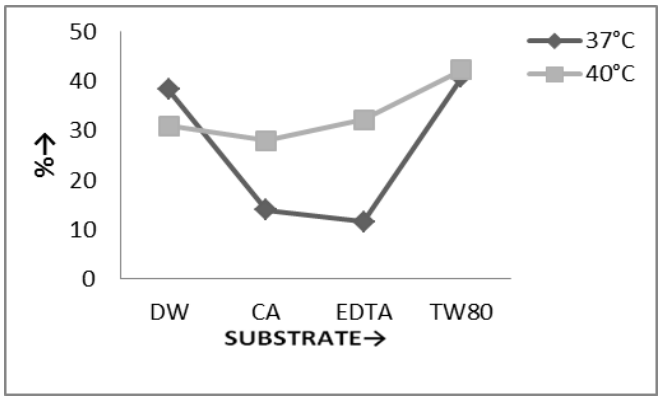

Fig. 2b- Removal of Chromium in Basal salt broth at pH 4 by viable cells.

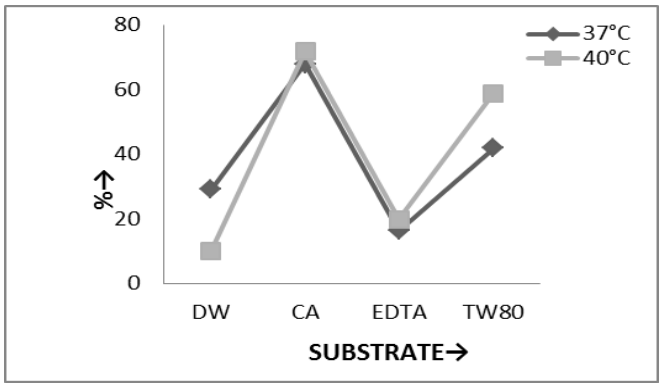

Fig. 2c- Removal of Chromium in Basal salt broth at pH 7 by Viable cells.

While at 40°C, 10.3% with distilled water, 72% with citric acid, 20% with EDTA and 59% with Tween 80 (Fig. 2c). At pH 9, percentage removal of chromium was 39.5% with distilled water, 14.2% with citric acid, 10.3% with EDTA and 69.86% with Tween 80 at room temperature (Fig. 2d). While at 40°C, 15.9% with distilled water, 14% with citric acid, 9% with EDTA and 65.4% with Tween 80 (Fig. 2d). At pH 12, Chromium was removed 19.2% in distilled water, 12.3% in citric acid, 19.3% with EDTA and 58.2% in Tween 80 at room temperature (Fig. 2e). While at 40°C, 33.3% with distilled water, 18.2% with citic acid, 17.1% with EDTA and 60.8% in Tween 80 solution (Fig. 2e).

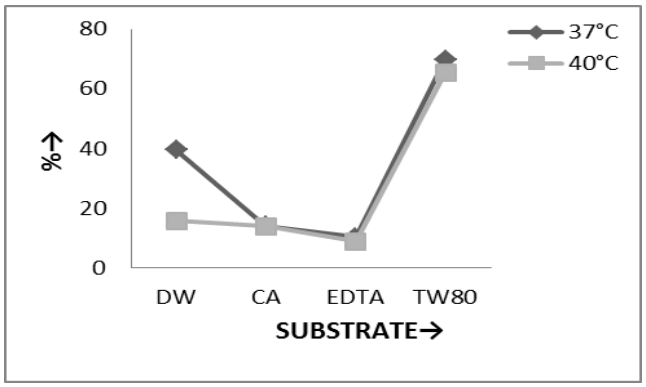

Fig. 2d- Removal of Chromium in Basal salt broth at pH 9 by Viable cells.

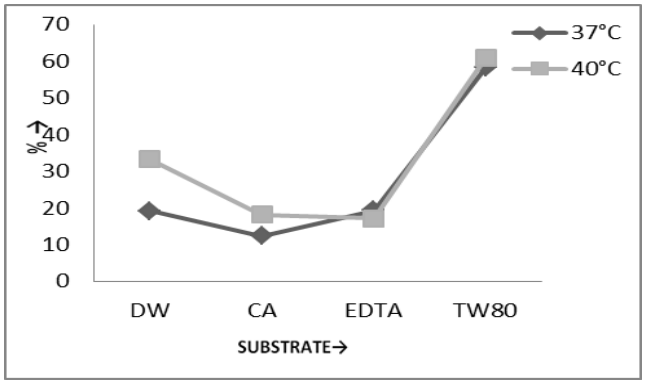

Fig. 2e- Removal of chromium in basal salt broth at pH 12 by viable cells

In Malt extract broth- At pH 2, Chromium was bioremediate 33.3% with distilled water, 20.77% with citric acid, 26.08% with EDTA and 40% with Tween 80 at room temperature (Fig. 3a). On the other hand at 40°C, with distilled water 22.8%, with citric acid 57%, with EDTA 18.52% and 77.3% with Tween 80 (Fig. 3a). At pH 4, Chromium was removed 47.3% with distilled water, 10.41% with citric acid, 17.33% with EDTA and 35.48% with Tween 80 at room temperature (Fig. 3b). At 40°C, the percentage removal of chromium was 27.4% with distilled water, 34.5% with citric acid, 26.6% with EDTA and 59.03% with Tween 80 (Fig. 3b). At pH 7, the percentage removal of chromium was 44.1% with distilled water, 10.34% with citric acid, 48.18% with EDTA and 22.85% with Tween 80 at room temperature (Fig. 3c). While at 40°C, 74.4% with distilled water, 20.01% with citric acid, 42.1% with EDTA and 44% with Tween 80 (Fig. 3c). At pH 9, the percentage removal of chromium was 57.7% with distilled water 17.85% with

citric acid, 1.8% with EDTA and 5.88% with Tween 80 at room temperature (Fig. 3d).

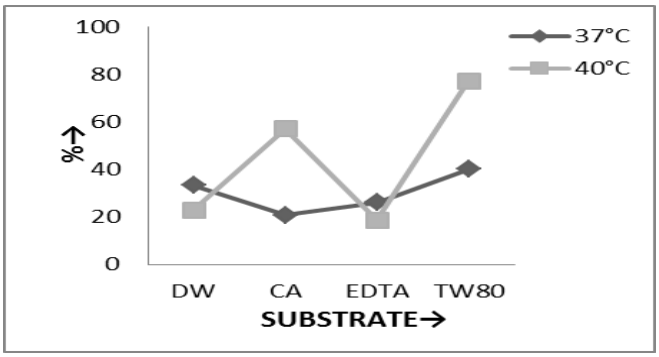

Fig. 3a- Removal of Chromium in Malt extract broth at pH 2 by Viable cells.

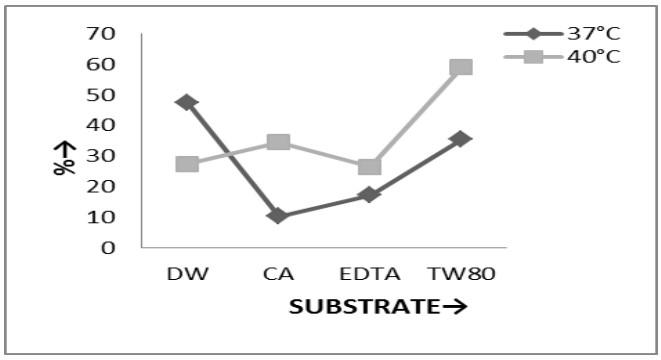

Fig. 3b- Removal of Chromium in Malt extract broth at pH 4 by viable cells.

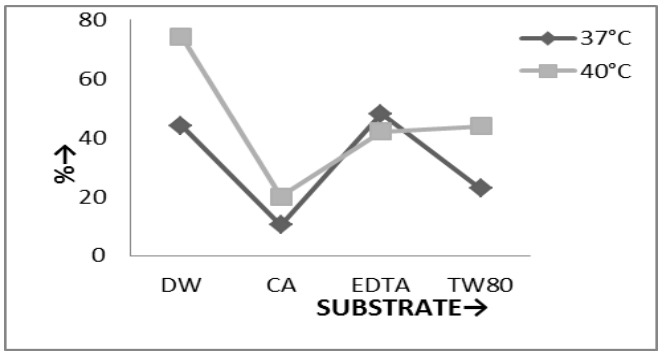

Fig. 3c- Removal of Chromium in Malt extract broth at pH 7 by Viable cells.

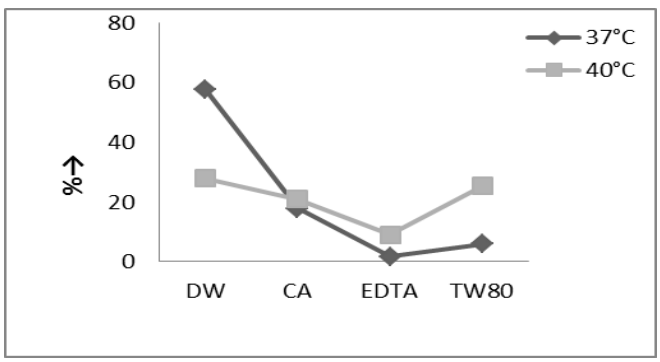

Fig. 3d- Removal of Chromium in Malt extract broth at pH 9 by Viable cells.

While at 40°C, 28% with distilled water, 21% with citric acid, 9.11% with EDTA and 25.3% with Tween 80(Fig.3d). At pH 12, percentage removal of chromium was 36.3% in Distilled water, 16.9% in citric acid, 12.8% in EDTA and 7.9% in Tween 80 at room temperature (Fig.3e). On the other hand at 40°C, 26.6% in distilled water, 26.5% in citric acid, 19.2% in EDTA and 28.5% in Tween 80 (Fig.3e).

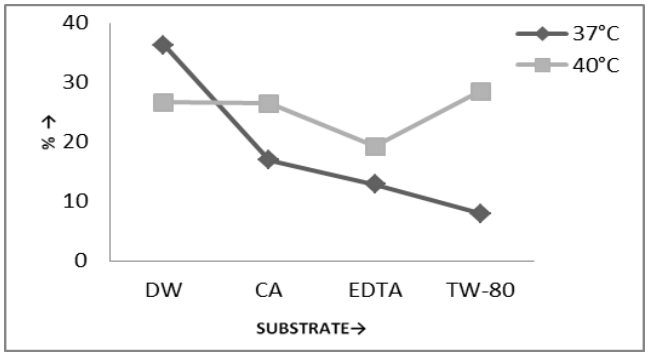

Fig. 3e- Removal of chromium in malt extract broth at pH 12 by viable cells

From the above findings it was cleared that at pH 2, viable cells in Tween 80 containing solution of malt extract medium showed better absorption of chromium (VI) at 400c then other fortified solutions. The reason behind this is at low pH due to the formation of more active sites on overall surface on fungal cell which increased adsorption and surfactant i.e. Tween 80 helpful in enhancement of chromium adsorption. In earlier studies, *Aspergillus* sp. N2 reduced 74% of Cr (VI) and *Penicillium* sp. N3 removed only 35% Cr (VI) at neutral pH. While in acidic pH *Aspergillus* sp. N2 was able to reduce 20% Cr (VI) from solution while *Penicillium* sp. N3 was reduce 93% of Cr (VI) from the solutions [32].

The physical and chemical research on the biosorbent demonstrated that the presence of polysaccharide and acidic functional groups on the fungus cell surface is more effective than the other parameters in other to the Cr (VI) ion removal.

Observation of Bioremediation at room temperature and 40°C with dried cells at different pH

In Basal salt broth- At pH 2, the percentage removal of chromium was 97.9% with distilled water, 82.20% with citric acid, with EDTA 80% and 97.42% with Tween 80 at room temperature (Fig. 4a). While at 40°C, 98.4% with distilled water, 85.3% with citric acid, 89.1% with EDTA and 98.2% with Tween 80 (Fig. 4a). At pH 4, Chromium was removed 94.2% with distilled water, 58.33% with citric acid, 55.5% with EDTA and 75.21% with Tween 80 at room temperature (Fig. 4b). While at 40°C, 99.5% with distilled water, 55% with citric acid, 68.2% with EDTA and 83.33% with Tween 80 (Fig. 4b). At pH 7, Chromium was removed 96.7% with distilled water, 45% with citric acid, 8.62% with EDTA and 94.04% with Tween 80 at room temperature (Fig. 4c). At 40°C, 99.71% with distilled water, 18.5% with citric acid, 10.6% with EDTA and 95% with Tween 80 (Fig. 4c). At pH 9, the percentage removal of chromium was 80.3% with distilled water, 17.9% with citric acid, 26.26% with EDTA and 42.1% with Tween 80 at room temperature (Fig. 4d). At 40°C, 98.7% with distilled water, 18.5% with citric

acid, 25.6% with EDTA and 62% with Tween 80 (Fig. 4d).

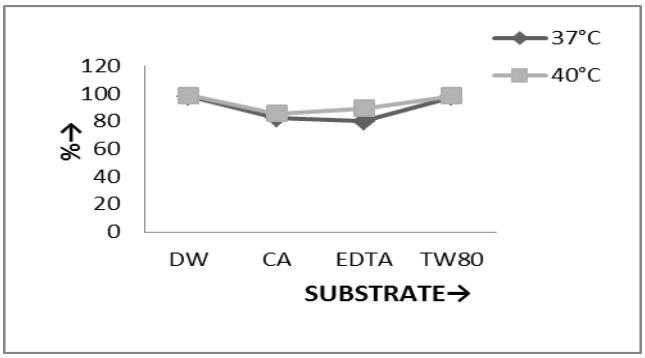

Fig. 4a- Removal of Chromium in Basal salt broth at pH 2 by Dried cells.

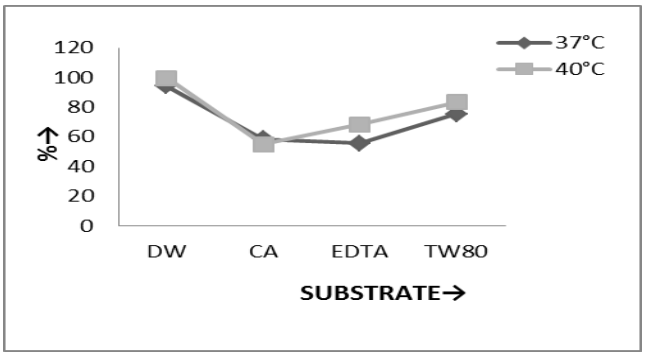

Fig. 4b- Removal of Chromium in Basal salt broth at pH 4 by Dried cells

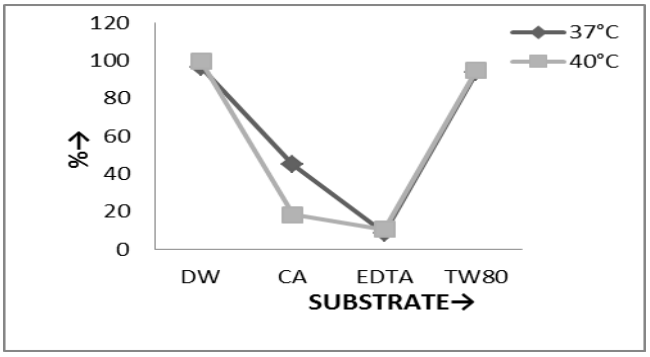

Fig. 4c- Removal of Chromium in Basal salt broth at pH 7 by Dried cells.

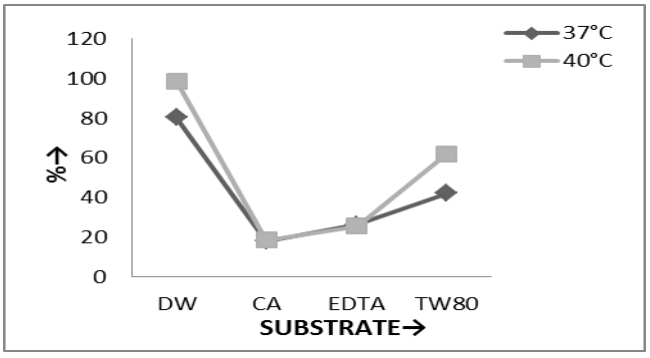

Fig. 4d- Removal of Chromium in Basal salt broth at pH 9 by Dried cells.

At pH 12, percentage removal of chromium was 82% in distilled water, 20.2% in citric acid, 30% in EDTA and 38% in Tween 80 at room temperature (Fig. 4e). while at 40°C, 97.1% in distilled water, 22.5% in citric acid, 26.7% in EDTA and 72.1% in Tween 80 solution (Fig. 4e).

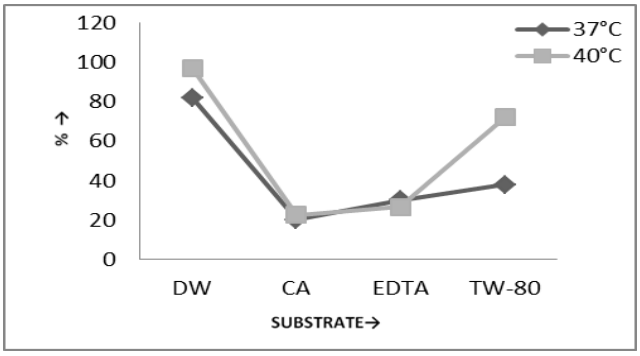

Fig. 4e Removal of chromium in Basal salt broth at pH 12 by Dried cell.

In Malt extract broth- At pH 2, Chromium was removed 66.6% with distilled water, 53.33% with citric acid, 36.3% with EDTA and 60.1% with Tween 80 at 40°C (Fig. 5a). while at room temperature with distilled water 12.5%, 53.52% with citric acid, 28.57% with EDTA and 82.5% with Tween 80 (Fig. 5a). At pH 4, chromium was removed 59.2% with distilled water, 12.76% with citric acid, 12.5% with EDTA and 90.9% with Tween 80 at room temperature (Fig. 5b). At 40°C, 78% with distilled water, 66.6% with citric acid, 16.5% with EDTA and 87.5% with Tween 80 (Fig. 5b). At pH 7, Chromium was removed 20.5% with distilled water, 44.18% with citric acid, 8.13% with EDTA and 76% with Tween 80 at room temperature (Fig. 5c). At 40°C, 75% with distilled water, 52.2% with citric acid, 18.9% with EDTA and 79% with Tween 80 (Fig. 5c). At pH 9, Chromium was removed 63.1% with distilled water, 43.9% with citric acid, 1.19% with EDTA and 99.1% with Tween 80 at room temperature (Fig. 5d). At 40°C, 80% with distilled water, 46.5% with citric acid, 8.19% with EDTA and 99.84% with Tween 80 (Fig. 5d). At pH 12, percentage removal of chromium was 24.4% in distilled water, 40.1% in citric acid, 5.9% in EDTA and 82.5% in Tween 80 solution at room temperature (Fig. 5e). While at 40°C, 20% in distilled water, 41.1% in citric acid, 12.7% in EDTA and 98.01% in Tween 80 solution (Fig. 5e).

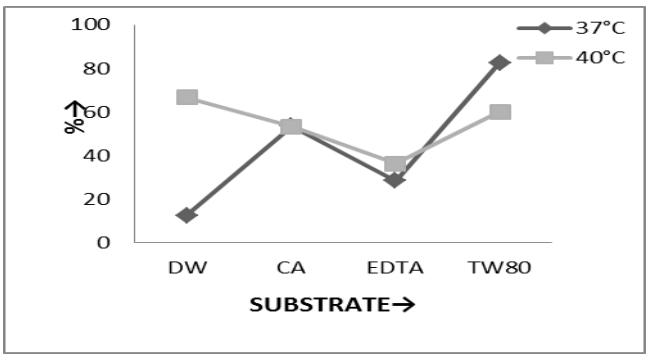

Fig. 5a- Removal of Chromium in Malt extract broth at pH 2 by Dried cells.

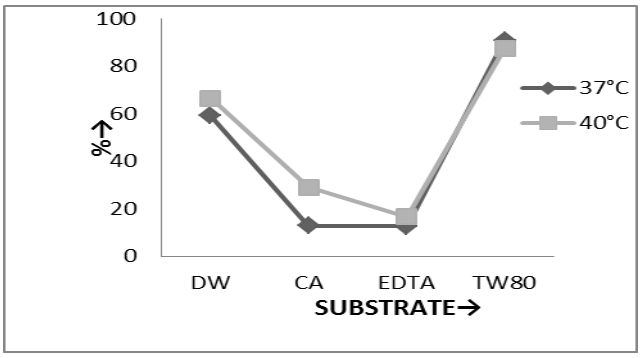

Fig. 5b- Removal of Chromium in Malt extract broth at pH 4 by Dried cells.

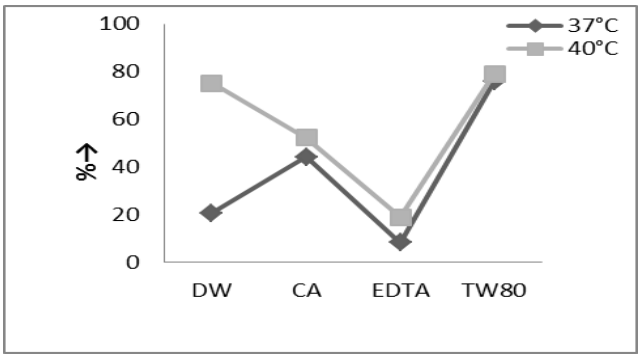

Fig. 5c- Removal of Chromium in Malt extract broth at pH 7 by Dried cells.

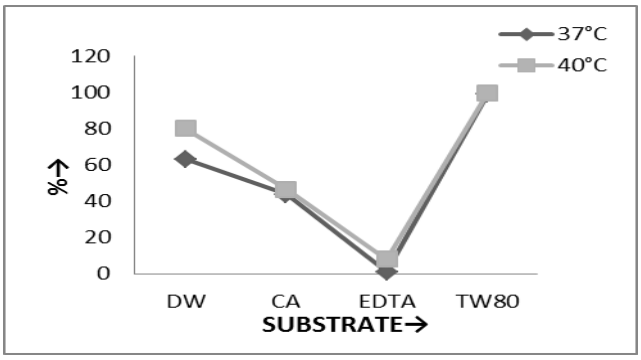

Fig. 5d- Removal of Chromium in Malt extract broth at pH 9 by Dried cells.

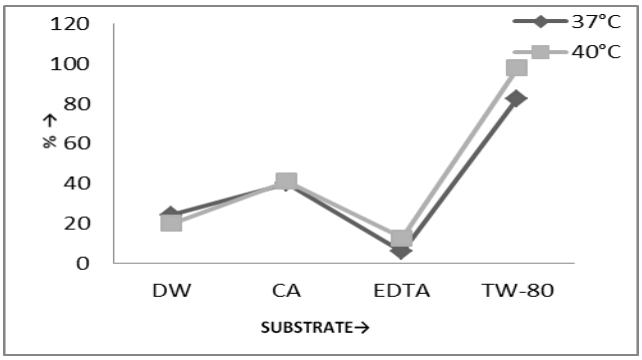

Fig. 5e- Removal of chromium in Malt extract broth at pH 12 by Dried cells

From the above results it was cleared that biosorbent cells shows better results at neutral and basic pH. In present study at pH 9, dried cells in Tween 80 containing malt extract agar medium shown better adsorption capacity then other fortified solutions at 400c temperature. And also these dried cells shown good results in distilled water at neutral pHs. Earlier, It has been also reported that biosorption capacities are pH sensitive and the biosorption increases as alkalinity of the solution increases [33]. Earlier, *Micrococcus luteus* also shows the same trend with copper and lead and the reason behind this was the presence of negative charge present on the fungal surface due to the functional groups which helpful in binding of heavy metals [34]. *Phanerochaete chrysosporium* fungus species has a higher adsorption capacity compared to the other microbial species due the spongy cell wall of the fungus.

Observations of Bioremediation at room temperature and 40° C with Immobilised cells at different pH

In Basal salt broth- At pH 2, the percentage removal of chromium was 24.7% with distilled water, 40% with citric acid, 31.03% with EDTA and 8.15% with Tween 80 at room temperature (Fig. 6a). On the other hand at 40°C, 14.4% with distilled water, 22.5% with citic acid, 41.2% with EDTA and 20.5% with Tween 80 (Fig. 6a). At pH 4, the percentage removal of chromium was 30.6% with distilled water, 35.5% with citric acid, 5.74% with EDTA and 3.33% with Tween 80 at room temperature (Fig. 6b). At 40°C, 57% with distilled water, 39% with citric acid, 12.7% with EDTA and 11.2% with Tween 80 (Fig. 6b). At pH 7, the percentage removal of chromium was 8.3% with distilled water, 27.27% with citric acid, 2.5% with EDTA and 67.39% with Tween 80 at room temperature (Fig. 6c). At 40°C, 26.7% with distilled water, 6% with citric acid, 12.5% with EDTA and 69.1% with Tween 80 (Fig. 6c).

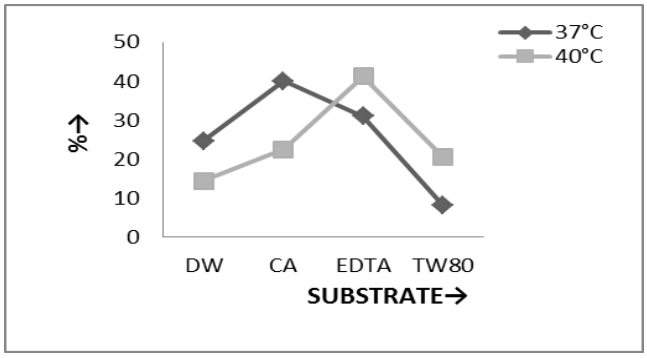

Fig. 6a- Removal of Chromium in Basal salt broth at pH 2 by Immobilised cells.

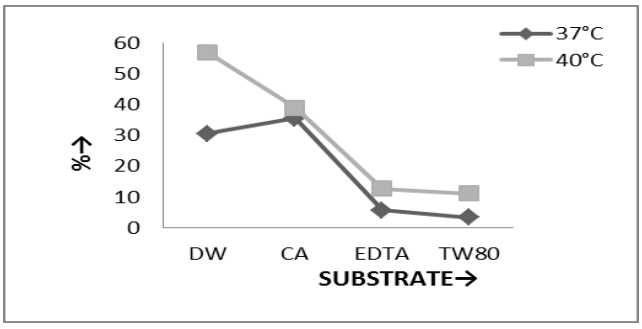

Fig. 6b- Removal of Chromium in Basal salt broth at pH 4 by Immobilised cells.

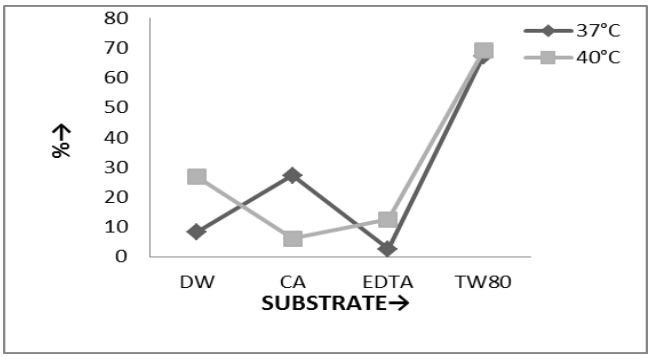

Fig. 6c- Removal of Chromium in Basal salt broth at pH 7 by Immobilised cells.

At pH 9, the percentage removal of chromium was 17% with distilled water, 30.61% with citric acid, 20% with EDTA and 23.01% with Tween 80 at room temperature (Fig. 6d). At 40°C, 39.03% with distilled water, 32% with citric acid, 29% with EDTA and 33% with Tween 80 (Fig. 6d). At pH 12, Percentage removal of chromium was 13.04% in distilled water, 42% in citric acid solution, 98.2% in EDTA and 19.1% in Tween 80 solution at room temperature (Fig. 6e). On the other hand at 40°C, 80.3% in distilled water, 29.1% in citric acid, 33.4% in EDTA and 28% with Tween 80 solution (Fig. 6e).

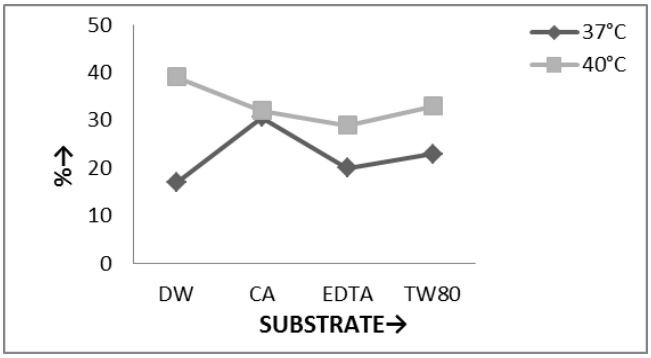

Fig. 6d- Removal of Chromium in Basal salt broth at pH 9 by Immobilised cells.

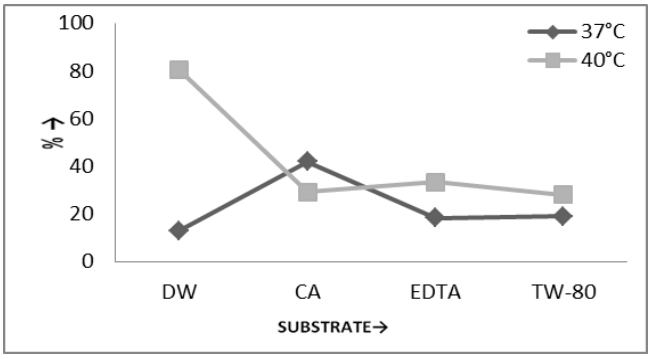

Fig. 6e- Removal of chromium in Basal salt broth at pH 12 by Immobilised cells

In Malt extract broth- At pH 2, Chromium was removed 80% with distilled water, 4.91% with citric acid, 1.88% with EDTA and 64.51% with Tween 80 at room temperature (Fig. 7a). While at 40°C, 75% with distilled water, 11.12% with citric acid, 8.11% with EDTA and 68.4% with Tween 80 (Fig. 7a). At pH 4, the percentage removal of chromium was 64.7% with distilled water, 12% with citric acid, 2.89% with EDTA and 29.41% with Tween 80 at room temperature (Fig. 7b). At 40°C, 53.3% with distilled water, 27.3% with citric acid, 7.3% with EDTA and 35.6% with Tween 80 (Fig. 7b). At pH 7, the percentage removal of chromium was 72.7% with distilled water, 19.23% with citric acid, 16.07% with EDTA and 20% with Tween 80 at room temperature (Fig. 7c). At 40°C, 57.1% with distilled water, 20% with citric acid, 15.5% with EDTA and 39.5% with Tween 80 (Fig. 7c). At pH 9, Chromium was 85.7% removed with distilled water, 11.11% with citric acid, 60.41% with EDTA and 94.13% with Tween 80 at room temperature (Fig. 7d). On the other hand at 40°C, the percentage removal of chromium was 44% with distilled water, 12.3% with citric acid, 68.4% with EDTA and 84.4% with Tween 80 (Fig. 7d).

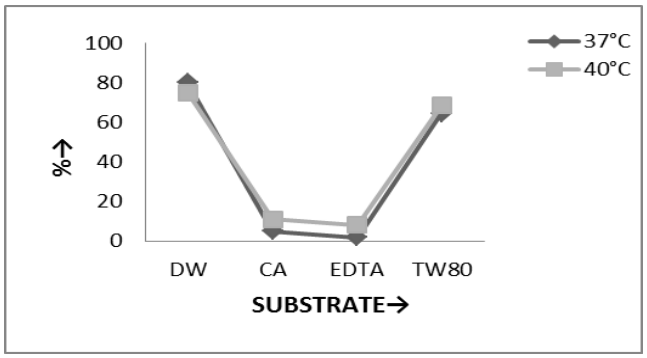

Fig. 7a- Removal of Chromium in Malt extract broth at pH 2 by Immobilised cells.

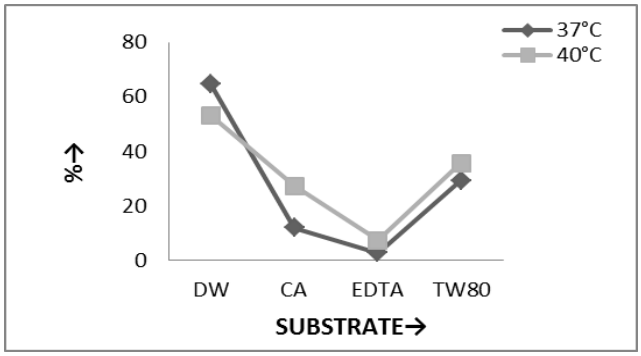

Fig. 7b- Removal of Chromium in Malt extract broth at pH 4 by Immobilised cells.

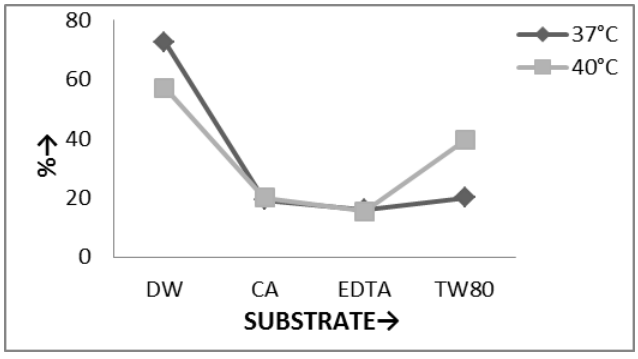

Fig. 7c- Removal of Chromium in Malt extract broth at pH 7 by Immobilised cells.

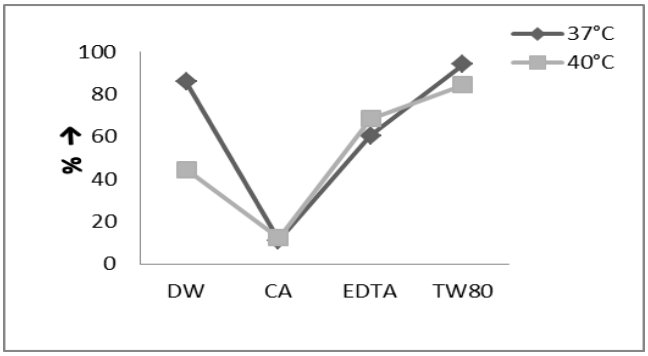

Fig. 7d- Removal of Chromium in Malt extract broth at pH 9 by Immobilised cells.

At pH 12, percentage removal of chromium was 46.1% in distilled water, 9.9% in citric acid, 65.2% in EDTA and 88.1% in Tween 80 solution at 40°C (Fig. 7e). While at room temperature, 27.2% with distilled water, 16.1% in citric acid, 55.2% in EDTA and 86.2% in Tween 80 solution (Fig. 7e).

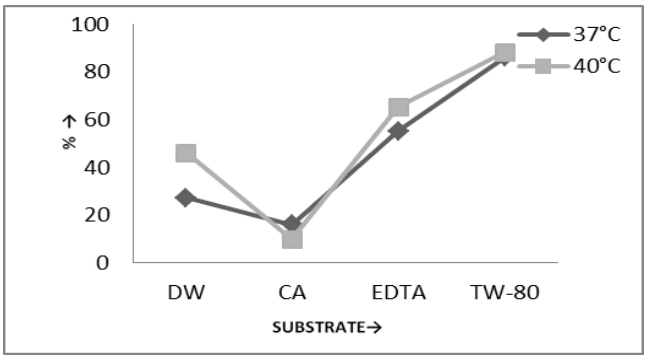

Fig. 7e- Removal of chromium in Malt extract broth at pH 12 by Immobilised cells

From the above results it was cleared that the immobilized cells showed abrupt results for the chromium adsorption. Immobilized cell at room temperature shown better adsorption of chromium at basic pH with basal salt medium containing EDTA. While in malt extract medium, immobilized cells shown good adsoption in basal salt medium containing Tween 80 at room temperature. In some results citric acid solution also had shown good results with immobilized cells.

In earlier studies various fungus like *Aspergillus niger* (Kumar and Bishonoi, 2008), *Penicillium Janthinellum* [35], *Rhizopus arrhizus* [36], *Aspergillus foetidus* [37], *Rhizopus nigricans* [38] shows 91.03%, 86.61%, 49.79%, 97%, 80% removal of chromium respectively. In one of the earlier study, the fungus, *Ganoderma lucidum* was used as a desorption of chromium under different substrate i.e. Distilled water, molasses, citrate, Tap water and EDTA and in this study EDTA shows maximum removal of chromium following molasses at pH 9 [39]. On the other hand, Tween 80 was used for the TNT mineralization by *Phanerochaete chrysosporium* and its shows better results as a enhancer substrate [40]. In the present study, the maximum removal was 99.7% with distilled water in basal salt broth at pH 7 under 40°C [49], with Citric acid was 85.3%at pH 2 in basal salt broth under 40°C, with EDTA was 89.1% at pH 2 in Basal salt broth under 40°C and with Tween 80 maximum removal was 99.84% at pH 9 in Malt extract broth under temperature 40°C. Here, pH seems to be the most important parameter in the biosorption process: it affects the solution chemistry of the metals, the activity of the functional groups in the biomass and competition between metallic ions [16]. The Biosorption of Chromium depends upon the ions surrounding the fungal cell during the uptake of metal ions. In present study, the viable cells, dried cells and immobilized cell of *Phanerochaete chrysosporium* shows the difference in metal uptake capacity at different range of pH and it is due to the ion exchange, electrostatic forces, chelation, adsorption by physical forces and chemical complexation due to the pH effects. White rot fungus *Phanerochaete chrysosporium* is a comes under category of crust fungus and the optimum temperature for growth for it is 40°C [41,42]. So, the variations in temperature shows the different results in case of *Phanerochaete chrysosporium.* A summary of compared results of present study with the earlier study shown in (Table 1).

Table 1- Bioremediation of chromium by several fungal spp and P. chrysosporium - a comparative study

Microorganism (Fungi)	Initial Conc. of Cr (VI)	Percentage removal of Chromium	References
Marine Aspergillus niger	400 mgL^{-1}	95.00	[43]
Aspergillus niger	30 mgL^{-1}	91.03	[35]
Aspergillus sydoni	30 mgL^{-1}	87.95	[35]
Penicillium janthinellum	30 mgL^{-1}	86.61	[35]
Aspergillus niger	10 mgL^{-1}	63.00	[44]
Rhizopus arrhizus	200 ppm	49.79	[36]
Aspergillus niger	500 ppm	75.00	[45]
Rhizopus arrhizus	100 mgL^{-1}	73.98	[46]
Aspergillus foetidus	5 mgL^{-1}	97	[37]
Rhizopus nigricans	100 mgL^{-1}	80	[47]
Aspergillus species	100 mgL^{-1}	92	[48]
Phanerochaete chrysosporium	100 mgL^{-1}	48.6	[49]
Phanerochaete chrysosporium	100 ppm	99.7	[50]
MTCC787	100 ppm	99.84 (Tween 80)	Present Study

Conclusions

From the study it was cleared that fungal dried cells (Biosorbent) is better then the other methodologies i.e. viable cells and immobilized cells and with Tween 80 fungal cells shows better biosorption to remediate the chromium from the synthetic solution in respect to the distilled water, Citric acid and EDTA. As per the earlier studies it is clear that this fungus showing better results then the other microorganism studied earlier for the bioremediation. In future aspects, *Phanerochaete chrysosporium* MTCC787 can be used to remediate the chromium contaminated sites and water bodies and it is an inexpensive and effective biomass to bioremediate the chromium.

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References

[1] Liao M., Chen C.L., Huang C.Y. (2005) *J. Environ. Sci.,* 17(5), 832-837
[2] Baath E. (1989) *Water Air Soil Pollut*., 47, 335-379.
[3] Doelman P. (1985) *Microbial Communities in Soil. Elsevier, London*, 369-384

[4] Duxbury T. (1985) *Advances in Microbial Ecology*, 8, 185-236.
[5] Tyler G. (1981) *Soil Biochemistry*, 5, 371-414.
[6] Patterson J.W. (1985) *Industrial wastewater treatment technology,* 2*nd ed*., 53-393.
[7] Shepherd C.M. and Jones R.L. (1971) *U.S. Clearing House Federal Science and Technology Information publication*, 71-7348.
[8] Schroeder D.C. and Lee G.F. (1975) *Water Air Soil Pollut*., 4, 355-365.
[9] Langand S. (1983) *Chromium: Metabolism and Toxicity*, 13-30.
[10] Petrilli F.L. and De Flora S. (1977) *Appl. Environ. Microbiol.,* 33, 805-809.
[11] Hueper W.C. (1966) *Occupational and environmental cancers of the respiratory systems*, 56-85.
[12] Mertz W. (1974) *Trace Elements Metabolism, Baltimore*, 2, 185-198.
[13] National Academy of Sciences (1980) *Drinking water and health safe drinking water committee*, 3, 364-369.
[14] Volesky B. (2001) *Hydrometallurgy*, 59, 203-216.
[15] Ahluwalia S.S. and Goyal D. (2007) *Bioresour. Technol*., 98, 2243-2257.
[16] Machado M.D., Santos M.S.F., Gouveia C., Soares H.M.V.M., Soares E.V. (2008) *Bioresour. Technol*., 99, 2107-2115.
[17] Garg U.K., Kaur M.P., Garg V.K. and Sud D. (2007) *Journal of Hazardous Materials*, 140, 60-68.
[18] Ranjan D., Srivastava P., Talat M., Hasan S.H. (2009) *Applied Biochemical Biotechnology*, 158, 524- 539.
[19] Gupta V.K., Rastogi A. (2009) *Journal of Hazardous Materials,* 163, 396-402.
[20] Mishara S.H., Doble M. (2008) *Ecotoxicology and Environmental Safety,* 71, 874-879.
[21] Kapoor A., Viraraghavan T., Cullimore D.R. (1999) *Bioresour. Technol*., 70, 95-104.
[22] Say R., Yilmaz N. and Denizli A. (2004) *Eng. Life Sci.*, 4, 276-280.
[23] Ahmad I., Zafar S. and Ahmad F. (2005) *J. Applied Sci. Environ. Mgt.*, 9, 123-126.
[24] Joshi P.K., Swarup A., Maheswari S., Kumar R. and Singh N. (2011) *Indian J. Microbiol.*, 51 (4), 482-487.
[25] Bumpus J.A. and Aust S.D. (1987) *PBC Essays,* 6, 166-170.
[26] Adenipekun C.O. and Fasidi I.O. (2005) *Afr. J. Biotechnol.*, 4, 796-798.
[27] Singh D. and Chen S. (2008) *Appl. Microbiol. Biotechnol.*, 81, 399-417.
[28] Bumps J.A., Tien M., Wright D. and Aust S.D. (1985) *Science*, 228, 1434-1436.
[29] Farrell R.L. (1987) *Philos. Trans. R Soc. London Ser. B,* 321, 549-553.
[30] APHA, AWWA and WEF (1999) *Standard Methods for the Examination of Water and Wastewater* 20*th Ed.*
[31] Churchill A., Walters T.V. (1996) *J. Environ. Eng.*, 121(10), 706-711.
[32] Tsubasa F., Yasuhiro I., Akane O., Tsutsumi K. and Morita H. (2008) *J. Gen. Appl. Microbiol.*, 54, 295-303.
[33] Zhang L., Zhao L., Yu Y.U., Chen C.Z., (1998) *Water Res.,* 32, 1437-1444.
[34] Yan G., Viraraghavan T. (2003) *Water Res.*, 37, 4486-4496.
[35] Kumar R., Bishonoi N.R. and Bishnoi K. (2008) *Chemical Engineering Journal*, 135, 202-208.
[36] Preetha B. and Viruthagiri T. (2007) *Sepration and Purification Technology*,, 57, 126-133
[37] Prasenjit B., Sumathi S., (2005) *J. Mater. Cycles Waste Manag., 7*, 88-92.
[38] Sudha Bai R., Emilia T., Abraham (2001) *Biores. Technol.,* 79, 73-81.
[39] Krishna K.R., Philip L. (2005) *J. Hazard Matter*., 121(1-3), 109 -117
[40] Hodgson J., et. al. (1987) *Canadian J. Microbiol.,* 46(2), 110-118.
[41] Burdsall H. (1985) *Mycologia Memoir*, 10, 61-63.
[42] Nakasone K. (1990) *Mycologia Memoir*, 15, 224-225.
[43] Khambhaty Y., Mody K., Basha S. and Jha B. (2009) *World J. Microbiol. Biotechnol*., 25, 257-260.
[44] Mungasavalli D.P., Viraraghavan T.H., Jin Y.C. (2007) *Colloids and Surfaces*, 301, 214-223.
[45] Srivastava S. and Thakur I.S. (2006) *Current Microbiology*, 53, 232-237.
[46] Prakasham R.S., Sheno Merrie J., Sheela R., Saswathi N., Ramakrishna S.V. (1999) *Environmental Pollution,* 104(3), 421-427.
[47] Sudha Bai R., Emilia T., Abraham (2001) *Biores. Technol*. 79, 73-81.
[48] Congeevaram S., Dhanarani S., Park J., Dexilin M. and Thamaraiselvi K. (2007) *J. Hazardous Materials*, 146, 270-277.
[49] Nikazar M., Davarpanah L. and Vahabzadeh F. (2008a) *Chemical Engineering Transactions*, 14, 475-480.
[50] Sumit Pal and Vimala Y. (2011) *Journal of Bioremediation and Biodegradation*, 2(5), 127.