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Bioremediation of Chromium by Viable cells, Biosorption and Immobilisation

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Chromium is the toxic pollutant present in industrial effluents. This pollutant is found to be present beyond the permissible limits i.e. 50 μg /ml. in zinc industry and the tanneries. The bioremediation of chromium was tested by the use of micro organisms by viable cell technique, biosorption and immobilization technique at room temperature and at 37°C . Eight microbial isolates were tested in the fortified solutions in the laboratory.

Key words: Chromium, Viable cells, Biosorption, Immobilization.

Chromium the 17th most abundant element in the earth's mantle. Its position in periodic table is with an atomic number 24 and mass number 51.9961, belongs to first series of transition metals. The elements vanadium, manganese and molybdenum surround its position in subgroup VIB of the periodic system. Its electronic configuration is (Ar) $3d^5 4s^1$.Chromium exists in a number of oxidation states of variable stability ranging from Cr(-II) to Cr(+VI) .Cr(III) is the most thermo dynamically stable species. these two oxidation states have different toxicity and mobility [Cr(VI)] a hexavalent carcinogen, via inhalation and is mobile, whereas trivalent chromium [Cr(III)] is comparatively less toxic and relatively immobile. Chromium is used in many industrial processes such as plating, alloying, tanning of animal hides, water corrosion inhibition, textile dyes, mordants, pigments, ceramic glazes, refractory bricks, pressure treated lumber, magnetic tapes, pigments for paints, cement, paper, rubber, composition floor covering and other materials. Its soluble forms are used in wood preservatives. Chromium is also the trace element responsible for the brilliant red and green colors of ruby and emerald. Chromium has attained wide public and regulatory attention

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because of its toxicity to the environmental ecosystems, including both microorganisms and animals under certain oxidation states. Elevated soil and water concentration of this potentially harmful element principally result from industrial wastes or spills. Tanning of leather with Cr salts [Cr (III)], first introduced in the year 1858 consumes considerable quantities of Cr.

Some of the earliest processes for chrome tanning of leather used Cr(VI)chemicals to saturate the skin and then reduce them to soluble forms. However the current standard practice now is to use a soluble trivalent compound and a masking agent, a procedure that allows Cr to effectively penetrate the hide. During this process Cr(III) forms cross links between the collagen fibers and gives leather its durable finish. The waste water discharged from tannery industry contains heavy loads of both organic and inorganic chemicals that are typically different from other Cr polluting industries. In natural systems, manganese oxides can oxidise [Cr(III)] to [Cr(VI)]. The maximum acceptable level in drinking water is 0.05 mg^{-L} (50μg^{-L})

Impact of Chromium on Environment

Humans exposure to hexavalent chromium salts for periods of 2 to 26 years has been implicated as a cause of cancer of the digestive tract. Bacterial populations resist to chromium as much as 500 $mg^{-L.}$ Fungal populations resist to Chromium as much as 500 mg^{-L}. Chromium hyper accumulator plant species Plant species such as *Sutera fodina, Dicoma niccolifera, Leptospermum scoparium* accumulate high concentrations of chromium in their plant tissues. Phytoextraction of chromium from contaminated tannery sludge.(*Scirpus lacutris, phragmites karka, Bacopa monnieri*)-able to absorb translocate and concentrate chromium in their tissues.

Chromium is the second most common metal found at sites, Industrial applications. Most commonly use chromium the hexavalent chromium [Cr)VI)] form which is acutely toxic and very mobile in ground water. Ground water extraction and treatment has traditionally been used to remediate chromium-contaminant plumes. Maximum contaminant goal level is MCLG is 0.1 parts per million (ppm) maximum contaminant level (MCL) MCL has also have been set at 0.1 ppm .

Health Effects

Short term effects include skin irritation or ulceration. Life time exposure at levels above the MCL, damage to liver, kidney, circulatory and nerve tissues and skin irritation.

The two largest sources of chromium emission in the atmosphere are from the chemical manufacturing industry and combustion of natural gas, oil and coal. When chromium is released to land, chromium compounds bind to soil are not likely to migrate to ground water. They are very persistent in water as sediments. There is high potential for accumulation of chromium in aquatic life. Treatment of tannery waste may be classified as physical, chemical and biological.In the physical treatment, the fill and draw sedimentation tanks are in use and found to be more efficient in the removal of suspended solids.About 98% of the chromium is precipitated in the primary sedimentation tanks and is removed along with the sludge. The sludge is dried over sand drying beds and can be used as a good manure. No appreciable education of dissolved solids, BOD, COD, color and chloride can be achieved in physical treatment process. But even with the primary treated tannery effluent lime encrustation occurs in the server, probably due to the precipitation of calcium carbonate and calcium sulphate. Chromium is present in the environment in several different forms.The most common forms are chromium (O), Chromium III and Chromium VI. Chromium is considered as an ultra trace mineral, since it is needed in such small quantity to perform its essential functions. The blood contains about 20 parts per billion (ppb) a fraction of microgram. Chromium helps the body to metabolize sugar, protein and fat by stimulating enzymes. Ingested large amount of Chromium (VI) can cause stomach upsets and ulcers, kidney and liver damage and even death. Long time exposure to chromium produced various effects in workers in the chromium industry. Workers occupationally exposed to chromium are considered to be risk for developing lung cancer. Limited information on the reproductive effects of chromium in humans exposed by inhalation suggests that exposure to chromium may result in complications during pregnancy and child birth (USEPA 1999) Acute animal test have shown chromium VI to have extreme toxicity from

inhalation and oral exposure, acute animal tests have shown chromium III to have moderate toxicity from oral exposure. USD HHS 1993.The normal range of whole blood chromium concentration is 2.0 ug/ml/100 ml to 3.0 ug /100 ml. (G. Prabhakaran, M. Mohan and N. Vijayalakshmi. 2005)Chromium bearing effluents has been reported through several methods such as reduction, precipitation, ion exchange, reaction with silica, electrochemical reduction, evaporation, reverse osmosis, and direct precipitation.Studies on the treatment of effluents bearing heavy metals have revealed adsorption to be highly effective, cheap and an easy method among the physico-chemical treatment processes. During to high cost and difficult procurement of activated carbon, efforts are being directed towards finding efficient and low cost adsorbent materials. A variety of low cost materials like fly ash, wood, charcoal, lignite coal, rice husk,, carbon, saw dust and ground nut husk have been tried. Activated carbons prepared from agriculture wastes also proved as efficient adsorbent materials.Adsorption capacities of activated carbons prepared from Mahua seed shells *(Madhuca indica)* for removal of CrVI. (C. Ramesh Babu, P. Raghunandam and K.N. Jayaveera 2004) Significant correlations were observed between diseases and heavy metals has been observed by (Rita Mehra and Meenu Junija. 2005) (i)skin disease and Cr. Mn, Fe, and Cu;(ii)Hypertension of cd, Mn, cu; (iii) Mental stress of cd, pb, Mn, Ni, cu, Zn; (iv)Diabetes and Cr, Mn, Ni; (v)Chest pain and pb; (vi)Respiratory trouble and Cr, Mn, Fe, Ni, Zn ; (vii)Tuberculosis and Zn ; (viii)Acidity and cd; (ix)Opthalamic problems and Mn, Fe, Ni and Zn .Removal of CrVI from aqueous Cr solutions was accomplished by using bacterial strains. The effect of PH(5,7 & 9) and contact time (24, 48, 72,96 hrs) were studied by conducting batch type experiments. The percentage metal removal has shown an excellent removal with increase in time. Irrespective of the concentration (10,20,30,40 and 50 ppm). Chromium was completely removed (100%) from the aqueous solution with the individual indigenous organisms. (M. Vasanthy and M. Sangeetha. 2004). Biosorption of metal

Table 1. Physicochemical and Microbiological analysis of effluents of Zinc industry

S. No	Parameter	Untreated effluent	Treated effluent	Permissible limit
1.	PH	8.47	2.75	6.5-7.5
2.	Conductivity(mMhos)	3.7	2.59	<2.5
3.	Turbidity (NTU)	4.3	1.7	5
4	Temperature (C)	30C	30C	28-30C
5.	MPN (coliform /100mL)	0.0	0.0	0-20
6.	TPC (cfu/mL)	4.7x106	2.0x104	<100
7.	D.O (mg/L)	3	0.0	5-8
8	TH (mg/L)	3457	>10000	75-150
9	CaH (mg/L)	2330	>1080	75-150
10	Mg H (mg/L)	1127	>90 20	75-150
11	Chlorides (mg/L)	199.6	>4991.4	250
12	Total Solids (mg/ L)	1800	36200	500-1000
13.	Total Dissolved solids (mg/L)	800	30400	500-1000
14	Suspended solids(mg/L)	1000	5800	500-1000
15	Nitrates (mg/L)	6.9	4.4	<45
16.	Ammonia (mg/L)	600	120	50
17.	Phenol (mg/L)	0.0	0.0	5.0
18.	Nickel (mg/L)	0.0	0.0	5.0
19.	Chromium(ug/ml)	80.0	50.0	5-10
20.	Copper (mg/L)	120.0	100.0	5.0
21	Zinc	100.0	20.0	5.0

ions by microbial biomass has long been recognized as an effective tool in bioremediation of industrial waste water. Dried mycelia biomass of 22 streptomyces isolates were screened for biosorption of hexavalent chromium [Cr(vI)] from aqueous solutions. The isolated were capable of removing 23-64% of chromium from a solution of 100 mg Cr(VI)/l in 24h. Streptomyces DBCC 747, the most potent isolate absorbed Cr(VI) rapidly during the early phase (8-12h) of incubation but continued steadily till 32h.The optimum concentration of biomass and Cr(VI) for bisorption were 15g/2 and 50 mg/2 respectively. Sorption of Cr(VI) was maximum at pH 2.0 and was affected adversely at neutral to alkaline PH, but increased gradually with increasing dehydration of the mycelia biomass. (Arundathi Pal and A.K. Paul, 2005). Hexavalent Chromium is a strong oxidant and it is toxic and carcinogenic to all forms of life extending from microorganisms

Table 2. Physicochemical and Microbiological analysis of effluents of tanneries

S. No	Parameter	Untreated effluent	Treated effluent	Permissible limit
1.	PH	8.1	8.0	6.5-7.5
2.	Conductivity(mMhos)	1.93	<2.5	<2.5
3.	Turbidity(NTU)	5.9	5	5
4	Temperature(C)	30C	30C	28-30C
5.	MPN Coliforms/100mL)	84	84	0-20
6.	TPC (cfu/mL)	8.8x109	1.8x108	<100
7.	D.O (mg/L)	0.0	0.0	5-8
8	TH (mg/L)	198	139	75-150
9	CaH (mg/L)	119	75	75-150
10	Mg H (mg/L)	116	94	75-150
11	Chlorides (mg/L)	222	159	250
12	Total Solids (mg/L)	1469	948	500-1000
13.	Total Dissolved solids(mg/L)	4329	2203	500-1000
14	Suspended solids(mg/L)	629	350	500-1000
15	Nitrates (mg/L)	47	30	<45
16.	Ammonia(mg/L)	171	40	50
17.	Phenol (mg/L)	0.0	0.0	5.0
18.	Nickel(mg/L)	0.0	0.0	5.0
19.	Chromium(ug/ml)	112	33	5-10
20.	Copper(mg/L)	11	0.4	5.0
21.	Zinc(mg/L)	0.0	0.0	5.0

Table 3. Bioremediation of hexavalent chromium: Percentage removal of chromium by viable microorganisms from synthetic solutions at room temperature after 24h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	48.0	53.0	54.0	45.9	91.0	41.0	72.95	38.9
4	53.4	55.0	60.1	48.0	89.0	46.0	90.59	42.4
6	68.0	61.5	65.8	59.0	87.5	53.0	91.8	58.9
7	72.0	85.5	82.0	65.0	93.0	55.0	98.82	55.3
8	64.6	75.0	70.5	58.0	86.0	51.0	81.82	0.0
10	71.3	75.0	803	62.0	89.0	54.0	84.8	65.9

BS-1= *Bacillus* species-1 ; BS-2 = *Bacillus subtilis;* PA= *Pseudomonas aeruginosa;* EC= *E. coli;* SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus*; SC= *Saccharomyces cerevisiae*; AN= *Aspergillus niger*

to animals. It is therefore apparent that there is an essential need to treat the industrial waste water for effective removal and reduction of toxic hexavalent Cr to relatively non toxic trivalent chromium.

Microbiological methods are now being used increasingly for the recovery and removal of metal ions from aqueous solutions. These applications vary from much smaller operations to large scale process and are competing well with

Table 4. Bioremediation of hexavalent chromium: Percentage removal of chromium by viable microorganisms from synthetic solutions at 37°C after 24h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	50.0	55.0	55.0	48.9	94.0	45.0	72.5	41.0
4	56.0	58.0	62.1	49.0	92.0	49.0	97.0	45.0
6	69.0	67.5	68.8	63.0	89.5	55.0	96.0	62.9
7	76.0	88.5	88.0	69.0	95.0	58.0	99.5	58.8
8	67.0	79.0	77.5	64.0	89.0	59.0	85.0	0.0
10	75.3	80.0	84.3	67.0	93.0	58.0	89.0	68.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

Table 5. Bioremediation of hexavalent chromium: Percentage removal of chromium by biosorption technique from synthetic solutions at room temperature after 3h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	27.0	13.0	41.0	25.9	41.0	10.0	42.0	18.0
4	8.0	15.0	16.0	18.0	39.0	13.0	40.0	22.0
6	24.0	21.5	23.0	28.0	37.5	15.0	41.0	38.0
7	26.0	35.5	10.0	34.0	43.0	10.0	48.2	35.0
8	21.6	25.0	28.1	26.0	36.0	11.0	31.0	0.0
10	12.0	25.0	48.1	35.0	39.0	14.0	34.8	39.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

Table 6. Bioremediation of hexavalent chromium: Percentage removal of chromium by biosorption technique from synthetic solutions at 37°C after 3h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	30.0	17.0	47.0	29.9	45.0	10.0	47.0	18.0
4	12.0	24.0	22.0	22.0	41.0	13.0	44.0	22.0
6	27.0	29.5	29.0	32.0	41.5	15.0	46.0	38.0
7	29.0	39.5	16.0	37.0	46.0	10.0	53.2	35.0
8	26.6	25.0	21.0	30.0	40.0	11.0	39.0	0.0
10	17.0	29.0	52.0	38.0	43.0	14.0	39.8	39.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

physical and chemical technologies. Biosorption of Cr VI by microbial biomass has already been recognized. The mechanisms associated with biosorption of heavy metals by microbial cells are complex and depend on the nature of physicochemical interactions of metal ions in the solution and extracellular as well as intracellular metal binding sites of the microbial cells. (Macaskie and Dean 1989). Inactivated and or thermally killed cells have been shown to accumulate metal ions to the same or greater extent than growing or resting cells. Microbial biomass of different types such as algae,filamentous Fungi yeasts have been used for uptake of Cr(VI) from aqueous solutions. Bacterial biomass derived from *Pseudomonas mendonica, Desulfovibrio desulfuricans. Thiobacillus ferroxidans*- effective tool for detoxification of Cr (VI) from aqueous solution. Different species of *Streptomyces* have been used for uptake and removal of trivalent chromium, lead, cadmium from aqueous solutions. Pollution due to chromium is one of the major problems of the present day. Chromium is discharged into the environment as result of anthropogenic activities involving steel manufacturing , wood preservation, leather tanning, dye and pigment, film and photography, metal cleaning, Chromium-VI a carcinogen cured in the production of steel, leather tanning, chrome plating and as a pigment in paints. When corporation's use chromium, it is convicted into Chromium VI and then they dump their waste into the environment Cr-VI can easily enter cells altering DNA, which blocks replication.

Difference between Chromium VI, and Chromium III is chromium III is an essential element for humans in trace concentration while,

Table 7. Bioremediation of hexavalent chromium: Percentage removal of chromium by biosorption technique from synthetic solutions at RT, after 6h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	35.0	21.0	50.0	34.9	50.0	10.0	53.0	25.0
4	17.0	27.0	28.0	28.0	49.0	13.0	49.0	29.0
6	29.0	33.0	35.0	39.0	50.5	15.0	53.0	43.0
7	32.0	41.0	28.0	41.0	55.0	10.0	53.2	40.0
8	29.6	29.0	32.0	38.0	49.0	11.0	45.0	0.0
10	22.0	30.0	58.0	43.0	43.0	14.0	43.8	45.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

Table 8. Bioremediation of hexavalent chromium: Percentage removal of chromium by biosorption technique from synthetic solutions at 37°C, after 6h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	40.0	31.0	60.0	44.9	60.0	10.0	63.0	35.0
4	26.0	37.0	38.0	38.0	59.0	13.0	59.0	39.0
6	39.0	43.0	45.0	49.0	60.5	15.0	63.0	53.0
7	42.0	51.0	38.0	51.0	65.0	10.0	63.2	50.0
8	39.6	39.0	42.0	48.0	59.0	11.0	55.0	0.0
10	32.0	40.0	68.0	53.0	53.0	14.0	53.8	55.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

if ingested, chromium VI is toxic, caring diseases of respiratory, circulatory and cancer Mixing strong acids and strong basis with Cr VI can lead to its remediation. House hold items containing acids and bases were mixed with chromium VI solution and tested to find the amount of Chromium VI can leads to its solution end tested to find the amount of Chromium VI in the system. For example : Hcl, Lemon Juice, white wine vinegar, coca cola, hydrogen peroxide, ammonia, sodium hydroxide (Clorox), ethylene glycol (antifreeze) well used to remediate Cr-VI was expected that ethylene glycol and strong acids like HCL and lemon juice work the best to remediate Cr.VI but Hcl did not work at all .NaoH and Ethylene glycol completely remediated Cr VI.Chromium (VI) is a known carcinogen that is very difficult to remove from groundwater. One potential method of remediation of chromium (VI) contaminated ground water involves using iron nano particles. The iron reduces chromium VI to chromium III which is less toxic and is cares to remove from ground water. Bench-top experiment have been performed to on. Well characterized solution in order to determine the kinetics of the reaction in order to determine the conditions under which the reaction is most effective. The studies have involved varying the PH of the solutions and the size of the nano-particles first order kinetics appears to describe the system. Experimental details and the results of these experiments are the subjects of this presentation. Kinetics of the remediation of chromium (VI)-contaminated groundwater using iron nano particles. Chitin and Chitosan are naturally abundant biopolymers which are of interest to research concerning the sorption of metal ions since the amine and hydroxyl groups on their

Table 9. Bioremediation of hexavalent chromium: Percentage removal of chromium by Biosorption technique from synthetic solutions at room temperature after 24h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	87.0	53.0	91.2	55.9	91.0	40.0	72.95	38.9
4	28.4	55.0	26.8	48.0	89.0	43.0	90.59	42.4
6	74.4	61.5	43.9	58.0	87.5	50.0	91.8	58.9
7	86.0	85.5	20.0	64.0	93.0	50.0	98.82	55.3
8	81.6	75.0	58.1	56.0	86.0	51.0	81.82	0.0
10	62.8	75.0	98.1	65.0	89.0	54050	84.8	65.9

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

Table 10. Bioremediation of hexavalent chromium: Percentage removal of chromium by biosorption technique from synthetic solutions at 37°C after 24h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	89.0	55.0	94.2	58.0	93.0	42.0	75.0	42.0
4	31.0	57.0	29.8	52.0	92.0	45.0	94.0	46.4
6	77.0	63.5	46.9	62.0	89.5	55.0	96.8	62.0
7	89.0	87.5	23.0	69.0	95.0	54.0	99.0	58.0
8	85.0	78.0	61.0	64.0	88.0	55.0	85.0	0.0
10	65.0	79.0	98.9	69.0	91.0	57.0	88.0	69.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

chemical structures as their chelation sites for metal ions.This study evaluates the removal of copper, chromium and arsenic elements from chromated copper arsenate (CCA) treated wool via biosorption by Chitin and Chitosan. Exposing CCA treated sawdust to various amounts of Chitin and Chitosan for 1,5 and 10 days enhanced removal of CCA components compared to remediation by deionised water only. Remediation with a solution containing 2.5 g chitin for 10 days removed 74% copper, 62% chromium, 63% arsenic from treated saw dust Samples using the same amount of chitosan as chitin resulted in 57% copper, 43% chromium and 30% arsenic removal. The results suggested that chitin and chitosan have potential to remove copper element from CCA treated wood. Thus there may be more abundant natural amino polysaccharides which could be important in the remediation of water wood treated with the newest formulations of organometallic copper compounds and other water borne preservatives containing copper. Agricultural soils receiving long term application of waste at Aligarh was analysed for the heavy metal content, diversity of metal tolerant filamentous fungi, and in vitro fungi-toxicity of heavy metals. The content of soil heavy metal was found more in treated soil as compared to untreated soil. The presence of common metal tolerant fungi recovered from soil includes the member of genera *Aspergillus, penicillium, Rhizopus, Fusarium, Trichoderma, Geotrichum Altenaria, Monilia* and other less frequently isolated fungi including mycelia and certain unidentified fungi. The total viable count of various groups of fungi ranged from 2.5 x 10^4 to 1.65 x 10^5 C.F.U./gm of soil. Heavy metal (Cr, Cu, Cd, Ni, Co) toxicity to fungi belonging to

Table 11. Bioremediation of hexavalent chromium: Percentage removal of chromium by immobilization technique from synthetic solutions at RT after 24h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	87.0	53.0	0.0	55.9	91.0	40.0	0.0	0.0
4	28.4	55.0	61.5	48.0	89.0	43.0	70.6	0.0
6	74.4	61.5	62.9	58.0	87.5	50.0	1.8	0.0
7	86.0	85.5	63.0	64.0	93.0	50.0	0.0	0.0
8	81.6	75.0	0.0	56.0	86.0	51.0	0.0	0.0
10	62.8	75.0	0.0	65.0	89.0	54.50	0.0	0.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

Table 12. Bioremediation of hexavalent chromium: Percentage removal of chromium immobilization technique from synthetic solutions at 37°C after 24h

pH	BS-1	BS-2	PA	EC	SA	SS	SC	AN
2	90.0	55.0	0.0	59.9	94.0	42.0	0.0	0.0
4	32.0	58.0	65.5	52.0	92.0	46.0	75.6	0.0
6	77.0	66.0	67.9	61.0	91.5	57.0	4.8	0.0
7	90.0	88.0	65.0	67.0	95.0	59.0	0.0	0.0
8	87.0	79.0	0.0	60.0	91.0	55.0	0.0	0.0
10	68.0	78.0	0.0	70.0	92.0	59.0	0.0	0.0

BS-1= *Bacillus species*-1 ; BS-2 = *Bacillus subtilis*; PA= *Pseudomonas aeruginosa;*
EC= *E. coli*; SA= *Staphylococcus aureus*; SS= *Staphylococcus saprophyticus;*
SC= *Saccharomyces cerevisiae;* AN= *Aspergillus niger*

genus *Fusarium Trichoderma, Geotrichum and Altenaria* and *Monilia* was evaluated *in vitro*. Conidial germination and growth of test fungi was observed upto 147 hrs at varying concentration (25-300 µg/ml) of metal in liquid medium. In the majority of cases conidial germination was delayed by 63 to 105 hrs of incubation and mat formation could not be detected even after 147 hrs of incubation in the isolated of *Geotrichum* and *Monilia.* On the basis of *invitro* toxicity, minimum fungicidal conc (MFC) of various metals was determined which ranged from <25 µg/ml to >2000µg/ml. The *Geotrichum* and *monilia* isolates were more susceptible to all metals as compared to genera like *Alternaria, Fusarium* and *Trichoderma*. Thus, fungi of metal contaminated soil have developed different levels of metal tolerance which may depend upon the nature of test fungi, metal toxicity and mechanism of metal fungi interaction. (Shaheem Zafar and Iqbal Ahmed, 2005).

The impact of tannery effluent from chingelpet district on various biochemical parameters viz. protein, carbohydrate and lipid content of various tissues such as muscle, liver ovary, skin, gill, brain, heart, intestine of larvivorous. Malaria control fish, gambusia affinis has been studied and found that there is maximum reduction in the protein content was observed in muscle form, and the carbohydrated content of was also found to decrease. Lipid content of muscle, liver, ovary, skin, gills, brain, heart and intestine was found to decrease to a lesser extent as compared to decline in protein and carbohydrate level of the tissues which was comparatively higher. (Revathi and Suma Gulati, Dawood Sharief 2005). Microorganisms resistant to both antibiotics and metals have been isolated from nosocomial and burn wound infections treated with metal contaminated environment such as estuaries, soil and sewage have suggested that the combined expression of antibiotic resistance and metal tolerance is resulting from metals present in an environment. Strains of *E.coli* are frequently resistant with heavy metal ions and the resistance can often be transferred by conjugation plasmid mediated resistance to mercury, cobalt, nickel, arsenate and arsenite has been observed in *E.coli.* some of these metals have been use in animal feeds for growth promotion or therapy. Others are known or suspected as causes of environmental pollution. Environmental toxicity of heavy metal to microorganism is affected by both the biotic and abiotic factors. These factors alter the bioavailability of metal ions.(Nilima Lankeswar and U.S Bacide 2005). The biological agents viz fungi and bacterial (*Pseudomonas sp)* were tried to develop a suitable technology for purification of industrial effluent. Pure culture of pseudomonal species and locally isolated fungi sp from mixed industrial effluents were utilized in the study. The pseudomonas sp. had shown capacity to grow in mixture of heavy metals (Cd, Co, Ni, Cr, Pb) of 5 ppm concentration of each element after 24 hrs of incubation.

The *Pseudomonas* sp and isolated fungi were subjected to different working parameters in order to find out the suitable conditions which favour the bioremediation process. The study indicated that bioremediation using pseudomonas sps could be effectively carried out under neutral pH value with 48 hrs to 72 hrs of contact time. The fungi sps was also effective in removal of heavy metals from mix industrial effluent having pH nearly neutral. The enhancement in the removal of heavy metals was quite remarkable with 72 hrs contact time compared to 48 hrs. The *pseudomonas sps* and locally isolated fungi from the effluent were found suitable for treatment purpose to purity the industrial waster water(.K.P. Patel , K.C. Patel, Swati J.Solanki and N.N Patel 2005). Chromium is found as picolinate or nicotinate and is used to help reduce body fat, increase muscle mass and boost energy levels.

Chromium is used in the body for carbohydreate metabolism and insulin function. The picolinate or nicotinate part of the supplement is chelated (bond) to other minerals such as calcium of B. Vitamins that aid intestinal absorption of Chromium which is usually low. Chromium picolinate. Daily ingestion of Chromium supplement of 600 micrograms for 5 years may lead to chromium accumulating in tissues at levels near those used in the study. To survive under metal-stressed conditions, bacteria have evolved several types of mechanisms to tolerate the uptake of heavy metal ions. These mechanisms include the efflux of metal ions outside the cell, accumulation and

complexation of metal ions inside the cell and reduction of heavy metal ions to a less toxic states (Nies 1999).

Minimal inhibitory concentrations

Heavy metals	MIC mM
Mercury	0.01
Silver	0.02
Gold	0.02
Chromium CrVI	0.2
Palladium	0.2
Platinum	0.5
Cadmium	0.5
Cobalt	1
Zinc	1
Thallium	1
Uranium	1
Canthamum	2
Uttrium	2
Scandium	2
Ruthenium	2
Aluminium	2
Lead	2
Iridium	2
Osmium	5
Antiomony	5
Indium	5
Rhodium	5
Gallium	5
Chromium CrII	5
Vanadium	5
Titanium	5
Beryllium	5
Chromium CrIII	10
Managanese	20

Mergeay *et al.,* (1985)

Tested MIC of several different metal ions for *E.Coli* on agar medium, and the toxic metal (lowest mic) was Mercury, whereas least toxic metal was manganese. Minimal inhibitory concentrations refers to the smallest concentrations necessary to inhibit growth, thus lower MIC values indicate more toxic metals and higher MICs indicate less toxicity.

RESULTS AND DISCUSSION

The results were tabulated from1-12 several industrial effluents were tested for the presence of chromium. The zinc and tannery industry showed the presence of chromium which is 80µg /l which is beyond the permissible limits i.e.50µg/l.The tanneries also showed chromium beyond the permissible limits i.e..112µg/l.

The bioremediation of Cr^{+6} by viable microorganisms (Table 3) showed a range of 0.0 - 98.8 % with *Saccharomyces cervisiae* the maximum % removal which was increased slightly when the microbes were (Table 4) incubated at 37°C. The bioremediation of Cr by biosorption technique (Table 5) for 3h contact period at RT showed 0.0 - 48.1 % with maximum removal by *Pseudomonas* at pH10. This was increased slightly at 37°C (Table 6) ranging from 0.0 - 52%, maximum by *Psedomonas* sp. The % removal of Cr after 6h of contact period at RT still increased the % removal (Table 7) of Cr than that of 3h contact period (0.0 - 5 8 %) which was further increased to 68 % at 37°C for 6h (Table 8). The % removal of Cr by biosorption technique after 24 h of contact period at RT (Table 9) showed 0.0 -9 8.82 % of Cr removal *Saccharomyces* with a maximum percentage removal at pH: 7. Similar experiments when conducted just by incubating at 37°C the percentage removal was slightly increased in responding microbes (Table 10) which showed a range of 0.0- 99 % Cr removal at pH: 7 & by *Saccharomyces cervisiae* . The Immobilization technique of Cr bioremediation when incubated at RT showed (Table 11) 0.0 - 93 % of Cr remova, l *S. aureus* with highest percentage removal at pH: 7. When similar experiments (Table 12) were conducted at different temperature, i. e 37°C there was slight increase in percentage removal of Cr with increase in temperature. It ranged from 0.0 – 95 %, *S. aureus* with a maximum percentage removal of Cr at pH 7.

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