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Biosorption of heavy metals from industrial waste water by *Geobacillus thermodenitrificans*

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ABSTRACT

The metal binding capacity of the thermophilic bacteria *Geobacillus thermodenitrificans* isolated from Damodar river, India was assessed using synthetic metal solutions and industrial waste water. Biosorption preference of dead biomass of *G. thermodenitrificans* for the synthetic metal solutions was in the following order $Fe^{+3} > Cr^{+3} > Co^{+2} > Cu^{+2} > Zn^{+2} > Cd^{+2} > Ag^{+} > Pb^{+2}$. It reduced the concentration of Fe^{+3} (91.31%), Cr^{+3} (80.80%), Co^{+2} (79.71%), Cu^{+2} (57.14%), Zn^{+2} (55.14%), Cd^{+2} (49.02%), Ag^{+} (43.25%) and Pb^{+2} (36.86%) at different optimum pH within 720 min. When this strain was applied in the industrial waste water biosorption preference was in the following order $Fe^{+3} > Cr^{+3} > Cd^{+2} > Pb^{+2} > Cu^{+2} > Co^{+2} > Zn^{+2} > Ag^{+}$ and concentrations reduced up to 43.94% for Fe^{+3}, 39.2% for Cr^{+3}, 35.88% for Cd^{+2}, 18.22% for Pb^{+2}, 13.03% for Cu^{+2}, 11.43% for Co^{+2}, 9.02% for Zn^{+2} and 7.65% for Ag^{+} within 120 min.

1. Introduction

Water is a valuable resource under increasing demand worldwide and is exposed to numerous sources of pollution. Industries such as mining, steel and electroplating, discharge aqueous effluents containing relatively high levels of heavy metals such as silver, cadmium, copper, cobalt, chromium, zinc, iron and lead. Untreated effluents from these manufacturing processes have an adverse impact on the environment [1–6] and remedial action is needed. A wide range of geological factors like sulphur deposition, global climate change, etc. and anthropogenic modifiers like acid wash of the metals in the electroplating industries, other heated discharges, riparian vegetation removal, flow modifications, etc. influences the thermal environment contributing increase in the concentrations of dissolved metals.

Physical and chemical methods can be used to remove metal ions from industrial effluent, but these are not commercially viable because of high operating cost or difficulty in treating the solid wastes generated [7]. Biosorption, bioprecipitation, and uptake by purified biopolymers derived from microbial cells provide alternative means of cleaning industrial effluents [7]. Various biomaterials have been examined for their biosorptive properties and different types of biomass have shown levels of metal uptake high enough to warrant further research [8]. Besides these biosorption utilizes various agricultural by-products such as, sugar-beet pulp [9], wheat bran [10], sugarcane bagasse [11] and natural materials including fungi, algae and yeasts [12,13].

Various naturally occurring bacteria exhibit high capacity for binding of metals. Intact microbial cells live or dead and their products can be effective bioaccumulators of both soluble and particulate forms of metals. Various microbial species under the genus *Bacillus* have been shown to be effective in bioaccumulation of chromium, iron, copper and other metal ions from polluted effluents both as immobilized cells and in the mobilized state [7,14,15].

Geobacillus thermodenitrificans can denitrify nitrate to nitrogen (gas). It was previously considered to belong to the species of *B. stearothermophilus* [16], now *G. stearothermophilus* [17].

The purpose of this study was to establish the ability of *G. thermodenitrificans* to biosorb and remove toxic metals from aqueous solutions by batch system.

2. Materials and methods

2.1. Sample collection site

Damodar is an important river of West Bengal, India and its tributaries are used to serve a variety of purposes including drinking, recreation, agriculture, and industry. The site, Kalajharia, is an important point-source of pollutants, where, mostly the untreated waste water of some steel industries, iron foundries and electroplating industries are released and drained into the Damodar river. Due to the ineffectiveness of purification systems, waste waters may become seriously dangerous, leading to the accumulation of toxic metal ions in the receiving water bodies with potentially

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Table 1
Physicochemical parameters of river Damodar at Kalajharia, West Bengal during March 2004 to February 2007.

Parameters	Maximum	Minimum	Mean	S.E.
BOD	12	4.8	7.75	0.37
COD	128	29.9	68.88	5.4
Dissolved O_2 (DO)	3.31	0.95	2.09	0.13
TDS	738	261.4	472.25	31.58
TSS	56.3	12.7	32.72	2.33
OG	319.9	232.6	278.58	4.35
Hardness	125.2	82.1	106.72	2.34
Alkalinity	281.3	118.9	182.63	8.83
Cl^-	118.5	46.8	84.55	3.28
F^-	1.43	0.14	0.85	0.07
NO_2^-	1.58	0.2	0.88	0.07
NO_3^-	8.78	0.35	2.65	0.51
Fe^{+3}	1.08	0.13	0.48	0.04
As^{+3}	0.11	0	0.03	0.01
Cu^{+2}	9.24	0.85	3.95	0.41
Cd^{+2}	0.73	0.02	0.3	0.05
Cr^{+3}	20.22	5.32	11.55	0.78

Data obtained from Ref. [20].

serious consequences on the ecosystem [18,19]. There are some of the important parameters which adversely affect the river water quality. Previous review of literature reveals that the point-source Kalajharia of the river Damodar is highly polluted [20] and some parameters indicating its pollution level is presented in Table 1.

2.2. Water sample collection

Sampling of the river water was done from Kalajharia industrial outfall. Grab samples of water were taken at a depth of 15 cm below the surface in triplicate and mixed to get a composite sample. Water was stored in high-grade plastic bottles of 2 L capacity. All the sample bottles were stored in iceboxes and brought to the laboratory for analysis. For microbiological analyses, 500 mL of water samples were taken from selected site employing aseptic techniques and stored in sterile Whirlpak type sealed plastic bags following standard collection and storage procedures. Aseptic techniques were observed during all microbiological analyses [20,21].

2.3. Isolation of bacterial strains from industrial waste water

The samples were sown in nutrient broth (Hi-media, Mumbai, India) and incubated at 65 °C for 48 h. The optimum temperature and pH for growth was 65 °C and 6.5, respectively. Pure culture was obtained by isolation of single colonies in nutrient agar (Hi-media, Mumbai, India) further purified by repeated streaking on agar plate. Isolated 30 colonies were investigated for their morphological characteristics and kept as slant cultures at 4 °C until use in further identification experiments.

2.4. Characterization and Identification of the isolated strains

To characterize the isolated bacterial strain, cells were grown at 65 °C in modified Luria Broth (mLB) medium [22]. The culture was sampled after 4, 12, 24 and 48 h intervals. The production of bacteria was monitored spectrophotometrically (Shimadzu, UV 1700, Asia specific), by measuring the absorbance at 600 nm [23].

The scheme of Cowan and Steel [24] was followed for characterization and identification of the strains. The results were interpreted using Bergey's Manual of Systematic Bacteriology [25]. The identification was confirmed by the Institute of Microbial Technology (IMTECH), Chandigarh, India.

2.5. Preparation of dead biomass of G. thermodenitrificans

The isolated strain was inoculated into 100 mL mLB in a 500 mL conical flask and incubated on a shaker at 100 rpm for 48 h at 65 °C. The cells were grown to late exponential phase, harvested by centrifugation (REMI, India) at 9000 rpm for 30 min at 4 °C and the pellet was washed three times with deionized water. Biomass concentrations of dead cell suspensions were determined by drying an aliquot in a preweighed aluminum foil container to constant weight at 80 °C [26,27].

2.6. Assessing the uptake of heavy metals from synthetic metal solution by dead biomass of G. thermodenitrificans

2.6.1. Preparation of synthetic metal solutions

Synthetic metal solutions were individually prepared by diluting 1000 mg/L of chromium (III) sulphate, iron (III) chloride, copper (II) sulphate, cadmium (II) chloride, silver (I) nitrate, zinc (II) sulphate, cobalt (II) nitrate and lead (II) nitrate solutions with deionized water to a desired concentration range between 25 and 175 mg/L. The initial concentration of the metals in the solution and samples after biosorption treatment were determined using an Atomic Absorption Spectrophotometry (AAS, PerkinElmer, model 2360).

2.6.2. Determination of optimum pH

In order to determine the optimum pH for biosorption of different metal ions by the bacterium, 50 mg of dead biomass of *G. thermodenitrificans*, were used in aqueous solution containing different metal ions (175 mg/L) with different pH values, ranging from 3 to 9, for 720 min on rotatory shaker (SI-300R, Korea) at 100 rpm. The solution pH was adjusted using 0.1N HCl and 0.1N NaOH to appropriate value before mixing the microorganism. Thereby, the necessary analysis was carried out.

2.6.3. Batch biosorption studies

Experiments with 8 different metal ions were conducted on 8 different days. For each metal ion various concentrations like 0 (control), 25, 50, 75, 100, 125, 150 and 175 mg/L were taken in conical flasks (150 mL) and mixed with 50 mg of dead biomass of *G. thermodenitrificans*, with optimum pH for various contact times (0, 60, 120, 180, 360 and 720 min) at 65 ± 1 °C and finally agitated (100 rpm) for 720 min on rotatory shaker (SI-300R, Korea). A control test without microorganisms was performed for each sample in parallel to avoid confusion between biosorption and possible metal precipitation. These experiments were done in triplicate for each metal ion side by side. The samples were finally sieved through a 14-mesh British Standard Sieve (BSS) and stored in plastic bottles at 4 °C to reduce the risk of the evaporation of the stored samples [28–30].

2.6.4. Metal uptake (q)

Biosorption of different metal ions using 50 mg of dead biomass of *G. thermodenitrificans* was carried in this study with 25 mL of each metal solution. Solution concentrations were varied from 25 to 175 mg/L and were agitated (100 rpm) on a shaker for 720 min which is more than ample time for adsorption equilibrium. Samples were taken at definite intervals for their final metal ion concentrations in the solution [31].

Metal uptake (q) was calculated using balance equation (Eq. (1)) [32]

$$q = \frac{V(C_i - C_f)}{1000} \times m \quad (1)$$

where q is milligram of metal ion biosorbed per gram of dead biomass (mg/g); C_i is initial concentration (mg/L); C_f is final con-

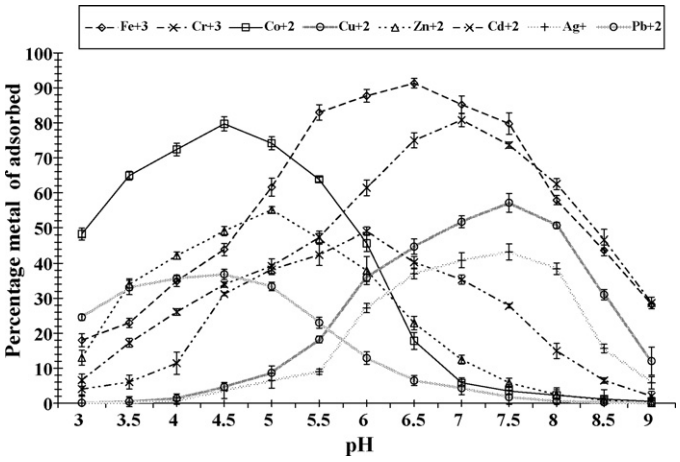

Fig. 1. Effect of pH on percentage of different metal biosorption by 50 mg dead biomass of *G. thermodenitrificans*.

centration (mg/L); V is volume of metal solutions in the flask (mL); m is weight of biosorbent (g).

Experiments were done in triplicate for each metal ion side by side and the average values were used in this study. Samples taken after proper incubation time period were analyzed for concentration of different metal ions using an Atomic Absorption Spectrophotometry (AAS, PerkinElmer, model 2360).

2.6.5. Equilibrium isotherm models

The Langmuir and Freundlich adsorption isotherms [33] are commonly used to model the biosorption studies based on metal concentration. The Langmuir equation is given by (Eq. (2)):

$$q_e = \frac{q_{max} K_L C_e}{1 + K_L C_e} \quad (2)$$

Langmuir equation transforms to the linearized form (Eq. (3)) [34].

$$\frac{1}{q_e} = \frac{1}{q_{max}} + \frac{1}{(q_{max} K_L) C_e} \quad (3)$$

where q_e is the metal ion sorbed (mg/g); C_e is equilibrium concentration of metal ion solutions (mg/L); q_{max} is maximum amount of metal ion which can be taken up by biosorbent (mg/g); K_L is Langmuir constant (L/mg).

The Freundlich equation is an empirical relationship describing the adsorption of solutes from a liquid to a solid surface. The general form of this model is:

$$q_e = K_F C^{1/n} \quad (4)$$

This can be linearized by taking logarithm of both sides of the equation to give:

$$\log q_e = \log K_F + \frac{1}{n} \log C_e \quad (5)$$

where K_f and $1/n$ are the Freundlich constants.

2.7. Assessing the uptake of heavy metals from industrial waste water by dead biomass of G. thermodenitrificans

2.7.1. Sample collection from industrial waste water

The method of water sample collection was same as described in Section 2.2.

2.7.2. Metal biosorption studies

Biosorption experiments were performed in triplicates at pH 6.5 (pH of the sampled industrial waste water) and contact time

Table 2
Frequency and morphological characteristics of the isolated bacteria from Kalajharia outfall.

Colony character	Number of isolated strain	% Frequency	Pigment	Gram stain	Cell shape	Cell arrangement	Cell size	Spore	Spore position	Code number	Name of the species	Strain number
Circular, convex, smooth, opaque	4	13.33	Cream	+ ve	Rods	–	2–4 μm	Present	–	BAC-1	*Bacillus mycoides*	MTCC 8340
Irregular, flat, moist, opaque	10	33.33	Cream	+ ve	Rods	Chains	2.5–5 μm	Present	Sub-terminal	BAC-2	*Geobacillus thermodenitrificans*	MTCC 8341
Circular, raised, smooth, opaque	1	3.33	–	+ ve	Rods	Single	2–4 μm	Present	–	–	–	–
Circular, raised, moist, opaque	1	3.33	Cream	+ ve	Rods	Pairs	2.5–4 μm	Present	Sub-terminal		–	–
Irregular, raised, smooth, opaque	1	3.33	–	+ ve	Rods	Single/pairs	2–4 μm	Present	Central		–	–
Circular, raised, lobate, opaque	2	6.66	Cream	+ ve	Rods	Pairs	2–4 μm	Present	Central	BAC-6	*Paenibacillus koreensis*	MTCC 8342
Circular, convex, smooth, opaque	2	6.66	Cream	+ ve	Rods	Chains	2.5–5 μm	Present	Central	BAC-7	*Paenibacillus larvae larvae*	MTCC 8343
Circular, pale, opaque	3	10.00	–	+ ve	Rods	Single/pairs and short chains	2–3 μm	Present	Central/sub-terminal	BAC-8	*Bacillus subtilis*	MTCC 8322
Circular, slightly raised, smooth and shiny, opaque	6	20.00	–	+ ve	Rods	Single/pairs and short chains	3–4 μm	Present	Terminal/sub-terminal	BAC-9	*Bacillus niacini*	MTCC 8323
	30	99.97										

Table 3
Biosorption equilibrium constant obtained from Langmuir, Freundlich isotherms for the biosorption of metal ions by dead biomass of *G. thermodenitrificans*.

Metal ions	Langmuir			Freundlich		
	q_{max} (mg/g)	K_L (L/mg)	R^2	K_f	$1/n$	R^2
Fe(III)	79.9	5.58	0.524	31.3	0.327	0.339
Cr(III)	70.7	2.09	0.746	10.4	0.533	0.805
Co(II)	69.76	2.22	0.675	27.16	0.257	0.726
Cu(II)	50.0	0.87	0.964	N.D.	3.88	0.957
Zn(II)	48.26	2.02	0.986	N.D.	5.16	0.963
Cd(II)	42.9	0.57	0.774	N.D.	8.15	0.695
Ag(I)	37.86	0.66	0.765	N.D.	11.61	0.729
Pb(II)	32.26	0.27	0.775	N.D.	5.88	0.782

N.D.—Not determined.

up to 120 min taking 25 mL of industrial waste water sample by 50 mg of dead biomass of *G. thermodenitrificans* following the same methodology described in Section 2.6.4.

2.8. Statistical analysis

The data obtained on the adsorption of different metals at different time interval, by the bacterium *G. thermodenitrificans* was subjected to *post hoc* Tukey test to analyze the differences in the rate of adsorption between the metals. Statistical analysis was done using SPSS ver 10 software [35,36].

3. Results and discussion

3.1. Isolation of the strain

Thirty morphologically distinct metal tolerant bacterial strains were screened from the study site for their metal tolerance limit (data not shown). Frequency of isolated colonies and their morphological characters are summarized in Table 2. All the isolates were Gram +ve rods [37]. The most frequent strains among the isolated bacteria were chosen and coded BAC-1 (13.33%), BAC-2 (33.33%), BAC-6 (6.66%), BAC-7 (6.66%), BAC-8 (10.00%) and BAC-9 (20.00%). Out of them only one isolate (BAC-2) was able to tolerate higher concentrations of different heavy metals.

3.2. Characterization and identification of the isolated strains

The isolated BAC-2 was a rod shaped, motile, endospore forming, Gram-positive, anaerobic bacterium and which was identified as *G. thermodenitrificans*. Morphological and physiological features and the biochemical profiles revealed that the colonies are irregular, flat; margins were rhizoid and off-white in color. Cells are Gram-positive rods 2.5–5.0 μm long with ellipsoidal sub-terminal spores and grow at 35–73 °C at pH 5–9 in 2–4% NaCl reduces nitrate to nitrogen gas; under anaerobic conditions. It is able to utilize glucose, fructose, galactose, maltose, mannose, cellobiose, D-xylose and L-rhamnosa as sole carbon sources. This strain is positive to catalase and oxidase reaction and negative to indole, methyl red and Voges–Proskauer test. It hydrolyzes starch and gelatin. Consequently, the recovered type strain has been deposited in the Microbial Type Culture Collection, IMTECH, Chandigarh, India Gene Bank under the accession number MTCC 8341.

3.3. Adsorption of heavy metals from synthetic metal solutions by dead biomass of G. thermodenitrificans

3.3.1. Effect of pH

Fig. 1 shows the effect of pH on the adsorption of different metals by *G. thermodenitrificans* dead biomass. Adsorption capacity (q) was analyzed over a pH range 3.0–9.0. It was apparent from the results that the metal adsorption was a function of pH; as the pH increased from 3.0 to 9.0, adsorption capacity increased at first for all the metals. In our experiments, metal adsorption by dead biomass of *G. thermodenitrificans* is very sensitive to pH with the maximum adsorption occurs at pH 6.5 for Fe^{+3}, 7.0 for Cr^{+3}, 4.5 for Co^{+2}, 7.5 for Cu^{+2}, 5.0 for Zn^{+2}, 6.0 for Cd^{+2}, 7.5 for Ag^{+} and 4.5 for Pb^{+2} respectively. The experiments beyond optimum pH for different metals were not carried as the metals precipitates at pH higher than optimum pH [38]. The maximum adsorption capacities (mg/g) for the different metals by dead biomass of *G. thermodenitrificans* were 79.9 mg/g for Fe^{+3} (at pH 6.5) > 70.7 mg/g for Cr^{+3} (at pH 7.0) > 69.76 mg/g for Co^{+2} (at pH 4.5) > 50.0 mg/g for Cu^{+2} (at pH 7.5) > 48.26 mg/g for Zn^{+2} (at pH 5.0) > 42.9 mg/g for Cd^{+2} (at pH 6.0) > 37.86 mg/g for Ag^{+} (at pH 7.5) > 32.26 mg/g for Pb^{+2} (at pH 4.5).

pH value is one of the main factors in biosorption efficiency by different organisms [39–44]. The different pH binding profiles for different metal ions are due to the nature of the chemical interactions of metal with the bacterial cells [30]. Solution pH influences cell surface metal binding sites and the interaction of these ions with bacteria may be primarily electrostatic in nature. pH affects the network of negative charges on the surface of the biosorbing cells (especially in case of Gram-positive bacteria) and the chemistry of the walls, as well as physicochemistry and hydrolysis of the metal [40,45].

3.3.2. Effect of initial metal concentrations

The percentage of adsorption was a function of the initial metal concentration. The amounts of metal uptake q (mg/g) by the dead biomass of *G. thermodenitrificans* at different metal concentration with time for different metals are shown in Fig. 2. The increase in initial metal concentration resulted in increase in the capacity of metal adsorption from 25 to 175 mg/L with a maximum adsorption at 175 mg/L for all the metals.

The maximum adsorption was observed for Fe^{+3} (91.31%) followed by Cr^{+3} (80.80%), Co^{+2} (79.71%), Cu^{+2} (57.14%), Zn^{+2} (55.14%),

Table 4
Results of *post hoc* Tukey test to confirm the differences in the rate of metal adsorption by *G. thermodenitrificans*.

	Cr^{+3}	Cu^{+2}	Co^{+2}	Cd^{+2}	Fe^{+3}	Pb^{+2}	Zn^{+2}
Cu^{+2}	33.078						
Co^{+2}	39.156	*6.078*[*]					
Cd^{+2}	18.489	51.567	57.644				
Fe^{+3}	*1.444*[*]	31.633	37.711	19.933			
Pb^{+2}	28.878	*4.200*[*]	10.278	47.367	27.433		
Zn^{+2}	43.578	10.500	*4.422*[*]	62.067	42.133	14.700	
Ag^{+}	49.244	16.167	10.089	67.733	47.800	20.367	*5.667*[*]

[*] Values in italics do not vary significantly at $P < 0.05$ levels.

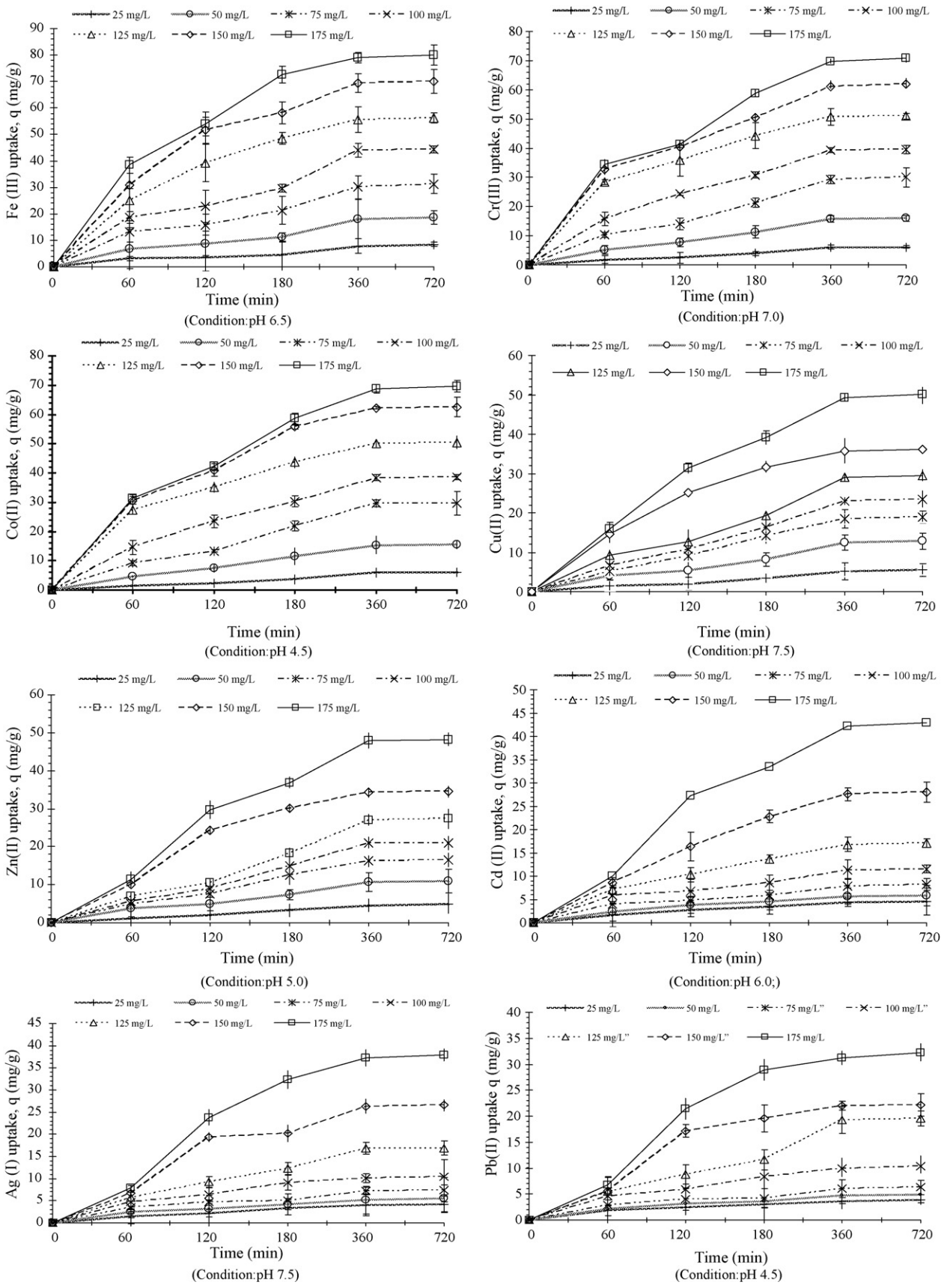

Fig. 2. Effect of initial metal concentration on biosorption of different metals by 50 mg/mL dead biomass of *G. thermodenitrificans* from synthetic metal solution (*condition*: biomass loading 50 mg, agitation 100 rpm).

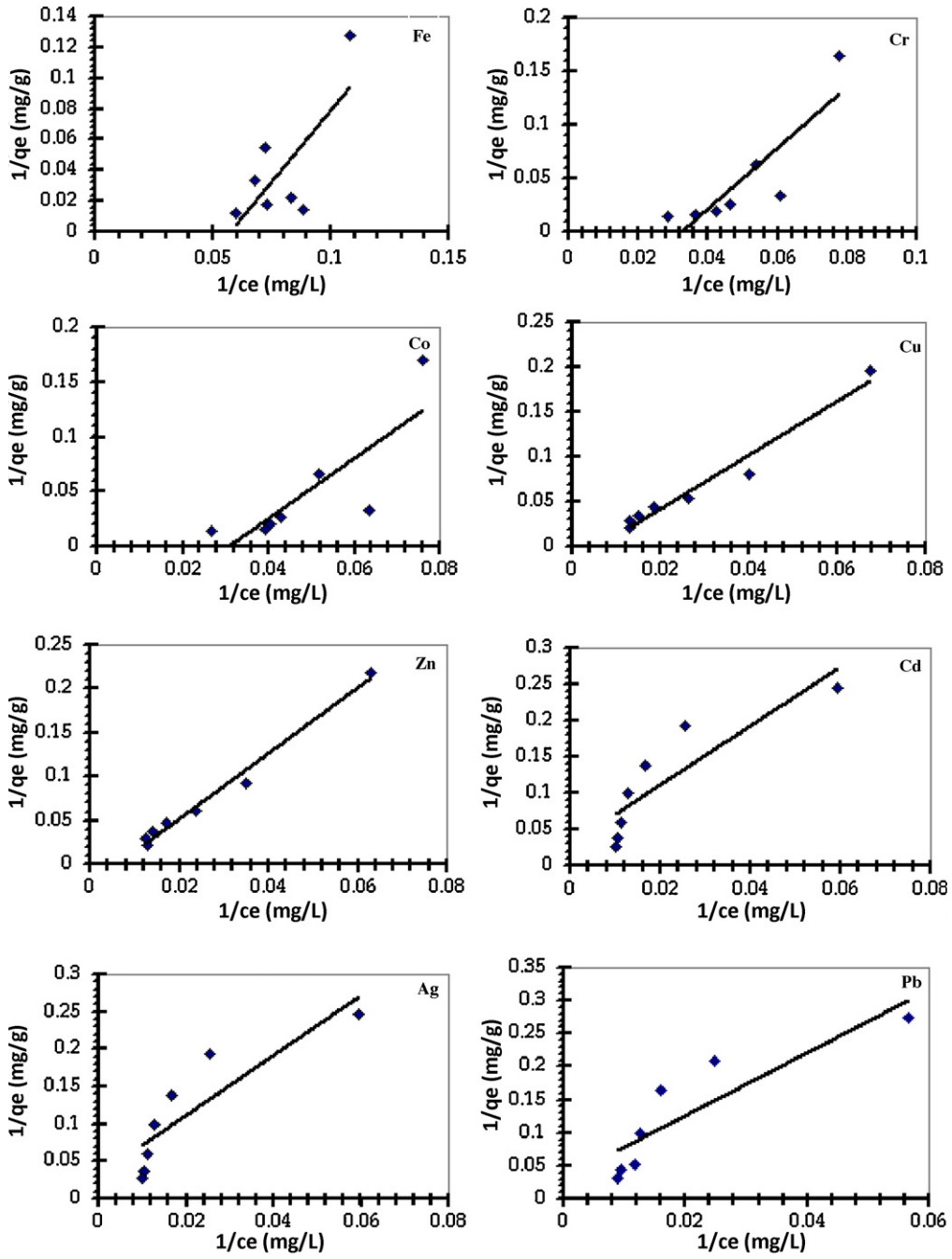

Fig. 3. Langmuir isotherm for different metal biosorption by *G. thermodenitrificans*.

Cd^{+2} (49.02%), Ag^{+} (43.25%) by dead biomass of *G. thermodenitrificans* and minimum adsorption was observed for Pb^{+2} (36.86%).

The enhancement in metal sorption could be due to an increase in electrostatic interactions, involving sites of progressively lower affinity for metal ions [46,47]. Macaskie [48] and Strandberg et al. [49] provided detailed description of mechanisms involved in metal microbe interactions. Tobin et al. [50] demonstrated that ions having a smaller ionic radius could be more quickly adsorbed onto a fixed area of adsorbent. The present study reveals that in the single metal solution the biosorption of Fe^{+3} was more than the other metals by the biomass. Ionic radius of Fe^{+3} (73.8 pm) was the smallest followed by Cr^{+3} (75.5 pm), Co^{+2} (83.8 pm), Cu^{+2} (87 pm), Zn^{+2} (88 pm), Cd^{+2} (109 pm), Ag^{+} (129 pm) and Pb^{+2} (133 pm). Ionic radius based higher biosorption of Fe^{+3} followed by other metals corroborated well with the findings of Cho et al. [51]; Pradhan and Rai [52].

The Langmuir (Fig. 3) and Freundlich (Fig. 4) adsorption isotherm are used to model the biosorption study based on metal ion concentrations. Langmuir model fitted better for Cu^{+2} and Zn^{+2} to experimental data in comparison to the model of Freundlich as represented from the value of its constants and R^2 (Table 3).

3.4. Adsorption of heavy metals from industrial waste water by dead biomass of G. thermodenitrificans

Microbial metal uptake by non-living cells, which is metabolism-independent passive binding to cell walls

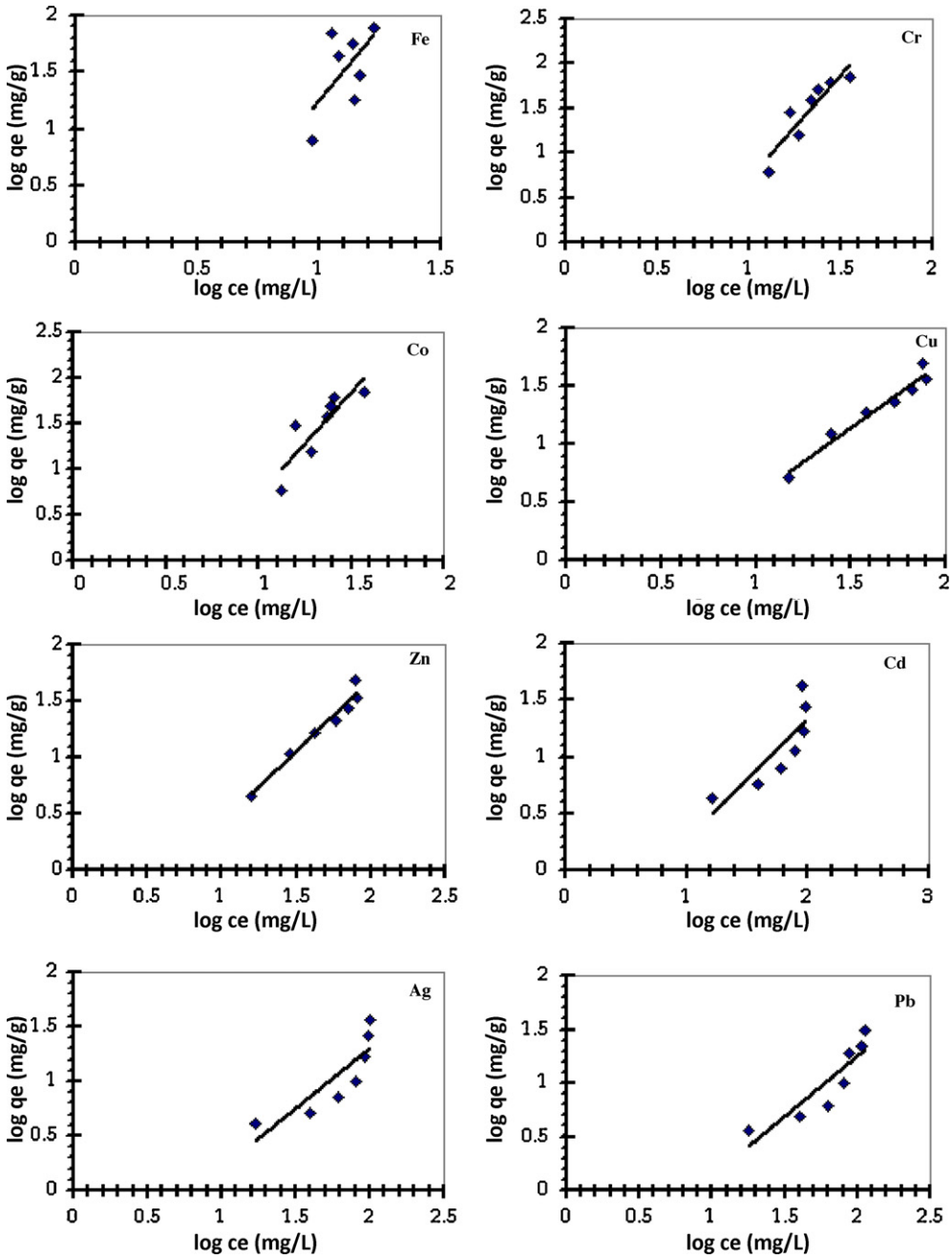

Fig. 4. Freundlich isotherm for different metal biosorption by *G. thermodenitrificans*.

(adsorption), and other external surfaces, is generally considered as a rapid process, taking place within a few minutes [30].

G. thermodenitrificans biomass absorbed Fe (III) ions from industrial waste water more readily than other ions (Fig. 5) within 120 min. Experiments indicated that sorption equilibrium reached much faster in case of industrial waste water samples (120 min) in comparison to synthetic metal solution (720 min) using same biosorbent. This may be due to the presence of co-metal ions in the industrial effluents [21]. The percentage sorption from industrial effluents was in the following order: $Fe^{+3} > Cr^{+3} > Cd^{+2} > Pb^{+2} > Cu^{+2} > Co^{+2} > Zn^{+2} > Ag^{+}$ and concentrations reduced up to 43.94% for Fe^{+3}, 39.2% for Cr^{+3}, 35.88% for Cd^{+2}, 18.22% for Pb^{+2}, 13.03% for Cu^{+2}, 11.43% for Co^{+2}, 9.02% for Zn^{+2}, 7.65% for Ag^{+} within 120 min.

3.5. Statistical analysis

The significant interactions between time and metals indicate that the adsorption varied with the amount of metals and the time interval. Thus at a specific time interval the amount adsorbed varied with the metal type which is evident from *post hoc* Tukey test (Table 4). However, the rate of adsorption was not significantly different for Cr^{+3}–Fe^{+3}, Cu^{+2}–Co^{+2}, Cu^{+2}–Pb^{+2}, Co^{+2}–Zn^{+2} and Zn^{+2}–Ag^{+} metal pairs.

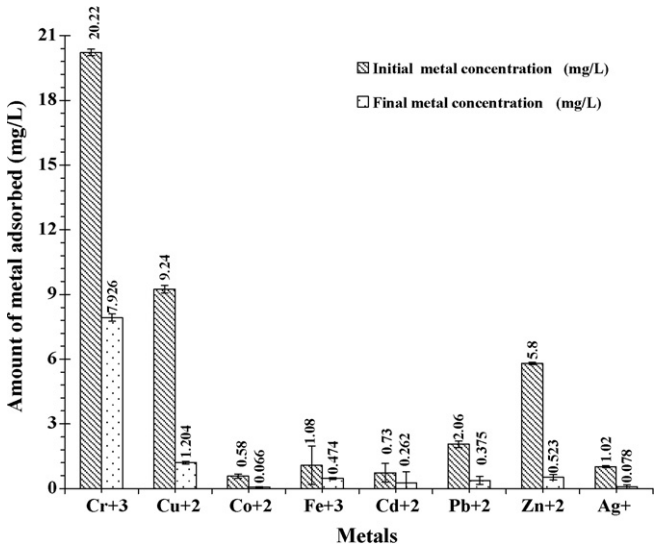

Fig. 5. Adsorption of metal ions from the industrial waste water by dead biomass of *G. thermodenitrificans* within 120 min.

4. Conclusions

The biosorption potentiality of heavy metals (Fe^{+3}, Cr^{+3}, Co^{+2}, Cu^{+2}, Zn^{+2}, Cd^{+2}, Ag^{+} and Pb^{+2}) by dead biomass of *G. thermodenitrificans* has been described for the first time. The results obtained indicate that initial metal concentration and pH highly affects the biosorption of heavy metals by *G. thermodenitrificans*. The metal adsorption by *G. thermodenitrificans* can be explained suitably by Langmuir adsorption isotherm. The present study concludes that isolated *G. thermodenitrificans* (MTCC 8341) strain may be used mainly for removal of Fe^{+3}, Cr^{+3} and Co^{+2} along with other heavy metals from steel and electroplating industry effluents.

Further study is necessary to find out the detailed mechanism of metal biosorption by *G. thermodenitrificans*.

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