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Distinct and effective biotransformation of hexavalent chromium by a novel isolate under aerobic growth followed by facultative anaerobic incubation

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Abstract A bacterial isolate (G161) with high Cr(VI)-reducing capacity was isolated from Cr(VI)-contaminated soil and identified as *Leucobacter* sp. on the basis of 16S rRNA gene sequence analysis. The isolate was a Gram-positive, aerobic rod. The hexavalent chromate-reducing capability of the isolate was investigated under three conditions of oxygen stress. The isolate was found to reduce Cr(VI) under all conditions but performed most effectively during aerobic growth followed by facultative anaerobic incubation. Under these conditions, the isolate tolerated $K_2Cr_2O_7$ concentrations up to 1,000 mg/l and completely reduced 400 mg/l $K_2Cr_2O_7$ within 96 h. The strain reduced Cr(VI) over a wide range of pH (6.0–11.0) and temperatures (15–45 °C) with optimum performance at pH 8.0 and 35 °C. The presence of other metals, such as Ca^{2+}, Co^{2+}, Cu^{2+}, Mn^{2+}, Ni^{2+}, and Zn^{2+}, induced no effect or else played a stimulatory role on Cr(VI)-reduction activity of the strain. The strain was tested for Cr(VI) removal in wastewaters and proved capable of completely reducing the contained Cr(VI). This is the novel report of a bacterial growth and Cr(VI)-reduction process under sequential aerobic growth and facultative anaerobic conditions. The study suggested that the isolate possesses a distinct capability for Cr(VI) reduction which could be harnessed for the detoxification of chromate-contaminated wastewaters.

Keywords *Leucobacter* sp G161 · 16S rRNA · Cr(VI) pollution · Aerobic growth · Facultative anaerobic Cr(VI) reduction

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Introduction

Chromium is widely used in manufacturing industries, e.g., electroplating, wood preservation, and steel production (Barnhart 1997). Since Cr(VI) is a powerful mutagen and carcinogen, the illicit disposal of Cr(VI)-containing wastewaters has raised serious environmental concerns (Ackerley et al. 2006). Conventional physico-chemical treatments for Cr(VI) removal are costly and are not completely effective (Kratochvil et al. 1998). Some microorganisms can reduce hexavalent chromium to the less toxic trivalent form, rendering them promising novel treatment agents of Cr(VI) pollution (Ganguli and Tripathi 2002; Kiran et al. 2007).

The Cr(VI)-reducing capacity of microorganisms has been reported under aerobic (Faisal and Hasnain 2004; He et al. 2009; Zahoor and Rehman 2009), anaerobic (Mabbett and Macaskie 2001; Romanenko and Korenkov 1977; Wang et al. 1989), or both conditions (Mclean and Beveridge 2001; Xu et al. 2011). The mechanisms involved in Cr(VI)-reduction have been elucidated. Under aerobic conditions, microbial Cr(VI) reductions are catalyzed by soluble chromate reductases in all chromate-reducing organisms except *Pseudomonas maltophilia* O-2 and *Bacillus megaterium* TKW3, which instead use membrane bound reductase (Blake II et al. 1993; Gu et al. 2006). A number of soluble chromate(VI) reductases have been purified and characterized, of which, two soluble Cr(VI) reductases, ChrR and YieF, were purified from *Pseudomonas putida*

MK1 and *Escherichia coli*, respectively (Ackerley et al. 2004a, b; Park et al. 2000, 2002). ChrR catalyzes Cr(VI) using a one-electron shuttle to form a short-lived intermediate Cr(V), followed by a two-electron transfer to produce stable Cr(III) (Ackerley et al. 2004b). Enzyme YieF catalyzes the direct reduction of Cr(VI) to Cr(III) through a four-electron transfer (Ackerley et al. 2004a). Under anaerobic conditions, both soluble and membrane-associated enzymes of the electron transfer system are reported to mediate Cr(VI) reduction. *Enterobacter cloacae* HO1 is resistant to chromate under both aerobic and anaerobic growth conditions, but Cr(VI) is reduced only by anaerobic cultures (Wang et al. 1989). Pattanapipitpaisal et al. (2001) reported that aerobic cultures of *Microbacterium* sp. MP30 grew well in the presence of 500 μM Cr(VI) but lost their Cr(VI)-reducing capability at such high concentrations. In contrast, under anaerobic conditions, Cr(VI) reduction to Cr(III) occurred, but growth was completely inhibited by exposure to 500 μM Cr(VI). All of these studies have contributed to our understanding of the Cr(VI) reduction process, but none have comprehensively explored combined bacterial growth and Cr(VI) reduction under varying conditions.

In the present study, a Cr(VI)-reducing bacterial strain was isolated from long-term chromium contaminated soil. The Cr(VI) reduction capability of the strain was investigated under aerobic growth followed by aerobic, facultative anaerobic, or strict anaerobic incubation. In addition, bacterial chromate-reduction efficiency was evaluated under varying conditions of temperature, pH value, initial Cr(VI) concentration, inoculum size, and the presence of other heavy metals. To assess the efficacy of the organism in practical situations, the ability of the strain to reduce Cr(VI) in wastewaters was investigated.

Materials and methods

Isolation and identification of the bacterial strain

A Cr(VI)-reducing bacterial strain was isolated from chromate-contaminated soil, located at a tannery factory in Wenzhou, China. The bacteria were maintained in Luria–Bertani (LB) medium comprising 10 g tryptone, 5 g yeast extract, and 10 g NaCl in 1 L distilled water, supplemented with 200 mg/l Cr(VI) in the form of $K_2Cr_2O_7$ with sterile $K_2Cr_2O_7$ stock solutions. The $K_2Cr_2O_7$ stock solutions were filter-sterilized through a 0.2 μm filter (Millipore, Bedford, USA) (Camargo et al. 2003a). Cultures were incubated at 35 °C with 160 rpm shaking. The strain was purified several times by streaking isolated colonies on fresh LB agar plates containing 1.5 % (w/v) agar. All chemicals were of analytical reagent grade.

The taxonomic studies of the isolates were analyzed using 16S rRNA gene sequencing. The universal bacterial primers 5′-AGA GTT TGA TCC TGG TCA GAA CGC T-3′ and 5′-TAC GGC TAC CTT GTT ACG ACT TCA CCC C-3′ (Yanagi and Yamasato 1993) were used in polymerase chain reactions (PCR). The reaction volume (20 μl) contained 10 μl 2× GC buffer, 20 μM of each dNTP, 10 pmol of each primer, and 1.5 U of Taq DNA polymerase. The PCR reactions were started with a denaturation step at 94 °C for 10 min, followed by 30 cycles, each performed denaturation at 94 °C for 30 s, annealing at 46 °C for 30 s, and elongation at 72 °C for 2 min with a final 10 min extension at 72 °C. Purified PCR products (Qiagen PCR Purification Kit) were cloned in a pGEMT vector and transfected into *E. coli* DH5α competent cells for sequencing. The obtained 16S rRNA gene sequence was compared with those deposited in GenBank using the BLAST program (Altschul et al. 1997). A phylogenetic dendrogram was constructed by the neighbor-joining method using MEGA-4.1 program (Saitou and Nei 1987). Sequence comparisons between strains were quantified using the Kimura-2-parameter distance model (Kimura 1980).

Cr(VI) reduction under different growth conditions

To prepare the seed culture for Cr(VI) reduction experiments, a purified colony of the Cr(VI)-reducing isolate was transferred to 10 ml LB medium plus 200 mg/l $K_2Cr_2O_7$ and incubated aerobically overnight on a rotary shaker (160 rpm) at 35 °C. To test the effect of oxygen stress on Cr(VI) reduction, 0.5 ml of the seed culture was transferred into 250-ml Erlenmeyer flasks containing 50 ml LB medium plus 400 mg/l $K_2Cr_2O_7$. The inoculated flasks were incubated for 48 h with agitation followed by an additional 48 h under either (a) agitation (aerobic) with 160 rpm shaking or (b) static facultative anaerobic conditions. For strict anaerobic conditions, cell suspensions were transferred to sterile, butyl rubber-sealed serum bottles degassed with O_2-free nitrogen and incubated for 48 h without shaking. Residual Cr(VI) and bacterial growth (OD_{600}) were determined on samples taken at predetermined intervals during the incubation period.

Cr(VI) analysis

One milliliter sample was withdrawn from the experimental liquid medium and centrifuged at 5,000×*g* for 5 min. The resulting supernatant was evaluated for Cr(VI) concentration. Cr(VI) removal was estimated as the decrease in Cr(VI) concentration in the supernatant. Amount of Cr(VI) was quantified spectrophotometrically at 540 nm following reaction with 1,5-diphenylcarbazide (Pattanapipitpaisal et al. 2001). The experiments were conducted in triplicate.

Other affecting factors

For testing the effect of pH on the reduction of Cr(VI) by the strain, autoclaved culture media were adjusted to 5.0, 6.0, 7.0, 8.0, 9.0, 10.0, and 11.0 with predetermined amount of filter-sterilized 1 M HCl or NaOH (Camargo et al. 2003a). Microbial Cr(VI) reduction was evaluated at six temperatures: 15 °C, 25 °C, 30 °C, 35 °C, 40 °C, and 45 °C. To assess the effect of inoculum size, Cr(VI) reduction was evaluated at inoculum volumes of 1 %, 5 %, 10 %, 20 %, 30 %, and 40 % log phase culture in LB medium containing 800 mg/l $K_2Cr_2O_7$. The Cr(VI) resistance and reduction ability of the strain was monitored at concentrations of 400, 600, 800, or 1,000 mg/l $K_2Cr_2O_7$. The effect of other heavy metals on Cr(VI) reduction was also investigated. The tested metal ions were Ag^+, Ba^{2+}, Ca^{2+}, Co^{2+}, Cu^{2+}, Mn^{2+}, Ni^{2+} and Zn^{2+}. All metal ions except Ag^+, were added in the form of chloride compounds (Xu et al. 2012), namely $BaCl_2$, $CaCl_2$, $CoCl_2$, $CuCl_2$, $MnCl_2$, $NiCl_2$, and $ZnCl_2$ were used. Ag^+ was added in the form of $AgNO_3$ (He et al. 2011). At the start of incubation, a final concentration 50 mg/l of each metal ion was added to the medium containing 400 mg/l $K_2Cr_2O_7$.

Cr(VI) in wastewaters removed by the isolated strain

Wastewater samples were collected from the ponds in the Lantian village of Longwan district and the industrial estate of Oubei district, Wenzhou City, China. The levels of $K_2Cr_2O_7$ contained in wastewaters were monitored (Table 1). The pH of samples 1 and 2 were evaluated at 4.57±0.17 and 5.95±0.28, respectively; these were adjusted to 8.0 with 1 M NaOH solution and 1 M HCl. Prior to dealing with the isolated bacterial culture, two samples were filtered through a coarse paper filter to remove sediments and then through 0.20 μm filters (Millipore, Bedford, USA) to capture the contained microorganisms.

Seed cultures (1 ml) grown aerobically overnight were transferred into 100 ml LB medium containing 400 mg/l $K_2Cr_2O_7$ in Erlenmeyer flasks and incubated at 35 °C with 160 rpm shaking. Following aerobic growth for 48 h, the bacterial cultures were harvested by centrifugation at 5,000×g for 5 min, and the pellets were suspended in wastewater samples of same volume as the original bacterial cultures. The suspended solutions were then incubated under aerobic, facultative anaerobic (without shaking), or strict anaerobic conditions. Residual Cr(VI) concentrations in the wastewater samples were analyzed as described above. The experiments were conducted in triplicate.

Accession numbers for nucleotide sequence data and the G161 strain

The obtained 16S rDNA nucleotide sequence data have been deposited in Genbank http://www.ncbi.nlm.nih.gov/nuccore/ under accession number GQ359816, and the strain *Leucobacter* sp. G161 has been conserved in the China Center of Industrial Culture Collection (CICC) http://www.china-cicc.org/ under accession number CICC10502.

Results

Identification of the Cr(VI)-reducing bacteria

After serial purifications, several bacterial colonies grew on LB agar plates containing 200 mg/l $K_2Cr_2O_7$, and one of these colonies was selected for further investigation. Physiological and morphological studies revealed the isolate as a short rod-shaped Gram-positive aerobe. Phylogenetic analysis revealed that the strain belongs to the genus *Leucobacter* and shares 99 % and 98 % similarity with *Leucobacter komagatae* strain IFO15245[T] (accession No.: D45063) and *Leucobacter albus* IAM 14851 (accession No.: AB012594), respectively, along the 1,488 bp sequence of 16S rDNA. The isolate was designated as *Leucobacter* sp. G161 and its 16S gene sequence was deposited in Genbank under accession number GQ359816.

Table 1 Cr(VI) content in wastewaters prior to treatment and after treatment with G161strain incubated under aerobic, facultative anaerobic, and strictly anaerobic conditions

Parameters	Wastewater 1		Wastewater 2	
	Cr(VI) content (mg/l)	Removal rate (%)	Cr(VI) content (mg/l)	Removal rate (%)
Before bio-treatment	5.46±0.53		9.53±0.71	
After bio-treatment				
Aerobically	0.92±0.18	83.2	2.83±0.64	70.3
Facultative anaerobically	0.12±0.00	97.8	0.35±0.02	96.3
Strict anaerobically	0.75±0.25	86.3	2.44±0.51	74.4

The potential Cr(VI) reduction ability and growth of the strain G161 under three different conditions

Cr(VI) reduction efficacy of the strain reared in LB medium supplemented with 400 mg/l $K_2Cr_2O_7$ under aerobic growth (first 48 h) followed by three different incubation conditions (second 48 h) was assayed as described in "Materials and methods" section. The strain was found to reduce Cr(VI) under all three conditions; however, complete reduction of 400 mg/l Cr(VI) occurred only in 96-h-old cultures incubated under facultative anaerobic conditions, while 33.3 % or 65.4 % of Cr(VI) was reduced under aerobic or strict anaerobic incubation, respectively (Fig. 1a). Continued aerobic incubation or changing to strict anaerobic conditions inhibited Cr(VI) reduction by the strain. The greatest propensity for Cr(VI) reduction was obtained under aerobic growth followed by facultative anaerobic conditions. This constitutes the first report of a bacterium that reduces Cr(VI) under these specific growth conditions.

Bacterial cells incubated aerobically (following 48 h aerobic growth) were slightly more numerous than cells subjected to facultative or strict anaerobic conditions post-48 h

(whose 96 h OD_{600} values under both conditions were essentially the same, Fig. 1b).

Cr(VI) resistance and reduction capability by the strain

Microbial Cr(VI) resistance and reduction were evaluated under aerobic growth for 48 h followed by facultative anaerobic incubation at varying concentrations of $K_2Cr_2O_7$ (0–1,000 mg/l). The strain tolerated $K_2Cr_2O_7$ concentrations as high as 1,000 mg/l; however, cell growth was markedly reduced at these levels (Fig. 2a). The effect of Cr(VI) concentration on Cr(VI) reduction efficacy is shown in Fig. 2b. Complete reduction of 400 mg/l Cr(VI) was observed at 96 h, while 85.1 % of 600, 49.0 % of 800, and 16.5 % of 1,000 mg/l Cr(VI) were reduced, respectively.

Other factors affecting Cr(VI) reduction

The effect of temperature on Cr(VI) reduction was evaluated. As shown in Fig. 3a, Cr(VI) reduction gradually increased within the temperature range 15–35 °C and decreased as temperature was raised higher. The Cr(VI)
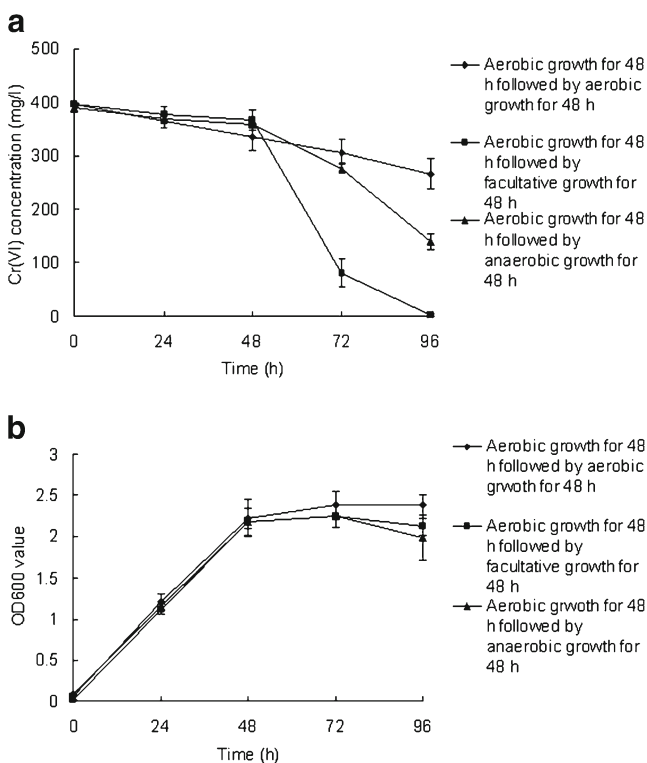

Fig. 1 Cr(VI) reduction and growth of the strain under three different growth/incubation conditions. Cr(VI) reduction (a) and growth (b) of the strain were measured at 24 h intervals during aerobic growth for 48 h followed by 48 h aerobic, anaerobic facultative, or strict anaerobic incubation. The cell growth was measured at OD_{600}. Values are mean numbers±standard deviations generated from three independent experiments
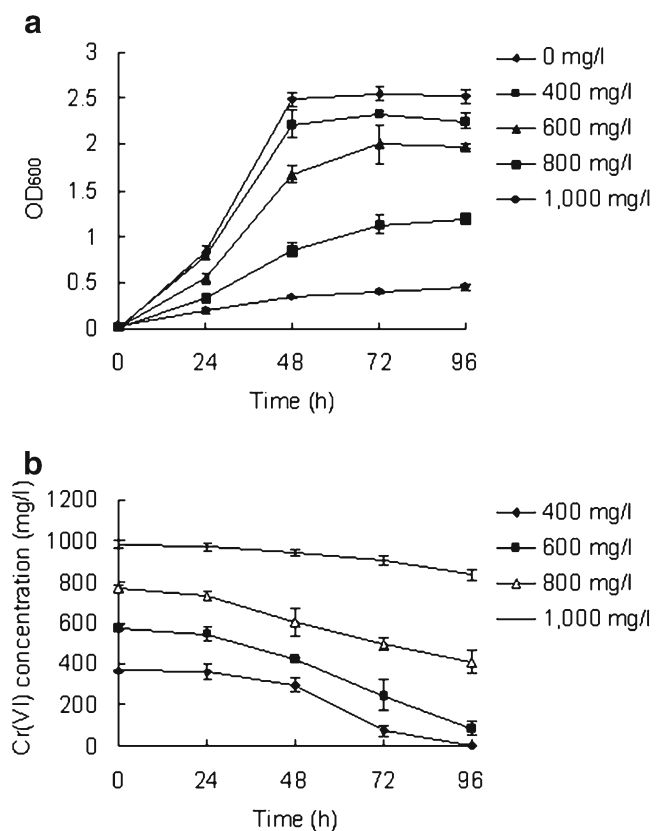

Fig. 2 Effect of Cr(VI) concentration on Cr(VI) reduction. The growth of the strain exposed to different concentration of Cr(VI) is assayed by OD_{600} (a), and the temporal effect of Cr(VI) concentration on Cr(VI) reduction is measured (b). Values are mean numbers±standard deviations generated from three independent experiments

Fig. 3 Effect of other parameters on Cr(VI) reduction. The effects of temperature (**a**), pH (**b**), bacterial inoculum volume (**c**) and heavy metal presence (**d**) on Cr(VI) reduction by the strain were estimated under aerobic growth for 48 h followed by anaerobic facultative incubation for 48 h. The amounts of Cr(VI) reduced at 35 °C (**a**), pH 8.0 (**b**), 40 % (**c**) and without the addition of metal ions (**d**) was set to 100 % as controls, the data at 15 °C, 25 °C, 30 °C, 40 °C, and 45 °C (**a**), pH 5.0, 6.0, 7.0, 9.0, 10.0, and 11.0 (**b**), 1 %, 5 %, 10 %, 20 %, and 30 % inocula (**c**) and with addition of metal ions (**d**) were normalized to the respective controls. Histograms display mean results±standard deviations generated from three independent experiments
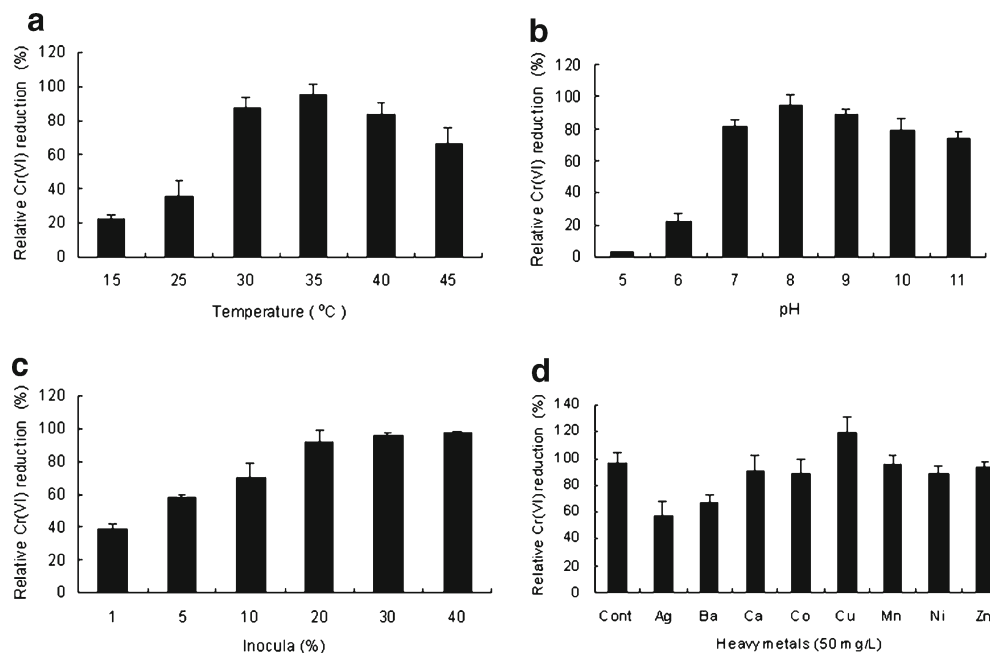
reduction was therefore optimal at 35 °C. Moreover, the Cr(VI)-reducing mechanism was strongly sensitive to lower temperatures, since less than 40 % Cr(VI) was reduced at 25 °C.

The effect of pH on the reduction of Cr(VI) is shown in Fig. 3b. Maximum Cr(VI) removal was observed at pH 8.0, and the strain was sensitive to acidic pH; only about 25 % of 400 mg/l Cr(VI) was reduced at pH 6.0. On the other hand, alkaline pH enhances the Cr(IV)-reducing performance of the strain.

Different inoculum volumes, ranging from 1 to 40 % log phase cultures, were also investigated. Cr(VI)-reduction by strain G161 increased as inoculum volume increased from 1 to 20 %, but no obvious further improvement appeared at higher inoculum volumes (over 30 %, Fig. 3c).

Since industrial processes may deposit other heavy metals besides Cr(VI) into wastewaters, the effect of metal ions on Cr(VI) reduction was evaluated. The ions assessed (namely, Ag^+, Ba^{2+}, Ca^{2+}, Co^{2+}, Cu^{2+}, Mn^{2+}, Ni^{2+} and Zn^{2+}) were separately added to the growth medium at concentrations of 50 mg/l. Addition of Cu^{2+} was found to stimulate Cr(VI) reduction, while Ag^+ and Ba^{2+} caused some decreases in Cr(VI) reduction ability, other ions exerted no obvious effect (Fig. 3d).

Cr(VI) of wastewaters removed by the strain G161

Finally, the potential efficacy of strain G161 as a bioremoval agent of Cr(VI)-contaminated wastewaters was assessed. Following 48 h bio-treatment by the strain, the contained Cr(VI) in wastewaters was removed by 97.8 % and 96.3 %, respectively, for the wastewater samples 1 and 2

under facultative anaerobic incubation (Table 1). Under aerobic and strict anaerobic incubation, the Cr(VI) in sample 1 was reduced by 83.2 % and 86.3 %, respectively, while the respective reductions were 70.3 % and 74.4 % for sample 2. These results demonstrate that aerobic growth followed by facultative anaerobic incubation is the most effective way to facilitate Cr(VI) reduction by the isolated strain.

Discussion

In the 1970s, it was first discovered that microorganisms in the genus *Pseudomonas* could reduce chromate and dichromate during anaerobic growth (Romanenko and Korenkov 1977). Since then, several additional chromate-reducing bacterial genera have been reported including *Bacillus* (Gu et al. 2006), *Enterobacter* (Wang et al. 1989, 1990), *Escherichia* (Ackerley et al. 2006), *Microbacterium* (Pattanapipitpaisal et al. 2001), *Ochrobactrum* (Faisal and Hsnain 2004; He et al. 2009), *Desulfovibrio* (Mabbett and Macaskie 2001), and *Pannonibacter* (Xu et al. 2011, 2012). However, few species in the genus *Leucobacter* have been comprehensively studied for their Cr(VI) reduction capabilities. Two new *Leucobacter* species, *Leucobacter chromiireducens* and *Leucobacter aridicollis*, have been isolated from activated sludge and demonstrated to be resistant to 4 μM Cr(VI), but the ability of these strains to reduce Cr(VI) was not further investigated (Morais et al. 2004, 2006). In this study, the strain G161 isolated from long-term Cr(VI) contaminated soil was identified as *Leucobacter* sp. based on 16S rRNA gene sequence analysis, and its Cr(VI)-reducing capability was studied.

Oxygen exerts a major effect on the growth and reduction efficacy of Cr(VI)-reducing bacteria. *Microbacterium* sp. MP30 fails to grow anaerobically, while aerobic growth is little affected by Cr(IV) concentrations as high as 500 μM Cr(VI); on the other hand, this species does not reduce Cr(VI) aerobically but does so under anaerobic conditions, even at 500 μM Cr(VI) (Pattanapipitpaisal et al. 2001). In the present study, the most effective Cr(VI) reduction was found to occur under aerobic growth followed by exposure to facultative anaerobic conditions. During the facultative anaerobic incubation stage, most of the bacterial cells settled near the bottom of the culture flasks, where oxygen was scarce. Oxygen has been previously reported to inhibit bacterial Cr(VI)-reducing activity (Wang et al. 1990). Consistent with this finding, more Cr(VI) was reduced under facultative anaerobic conditions than under aerobic conditions, following aerobic growth for 48 h in the present study. Under strict anaerobic conditions, the change from oxygen-filled to nitrogen-filled anaerobic environment forces rapid adjustment in bacterial metabolism. Changes in enzyme production result in decreased chromate(VI) reduction under strict anaerobic (relative to facultative anaerobic) incubation. Combining growth with incubation, aerobic conditions encourage growth of the strain, while the facultative anaerobic phase facilitates effective Cr(VI) reduction.

Bacterial chromate reduction and resistance are independent processes (Ohtake et al. 1987). In previous studies, an *Ochrobactrum* sp. strain CSCr-3 from chromium landfill could tolerate up to 800 mg/l Cr(VI) and had completely reduced 100–200 mg/l Cr(VI) at 96 h incubation (He et al. 2009). A *Pseudomonas* strain CRB5 had completely reduced 20 mg/l Cr(VI) following 120 h incubation (McLean and Beveridge 2001). By comparison, the G161 strain performed considerably better. The strain tolerated Cr(VI) concentrations up to 1,000 mg/l and had completely reduced 400 mg/l Cr(VI) within 96 h (Fig. 2a, b). Temperature is a critical factor in microbial Cr(VI) reduction. The optimal temperature for Cr(VI) reduction by G161 was found to be 35 °C. The same optimal temperature was reported in previous observations of Cr(VI) reduction by *Ochrobactrum* sp. strain CSCr-3 (He et al. 2009). Cr(VI) removal was favored under alkaline conditions, with optimum pH value for Cr(VI) reduction being 8.0, consistent with that of other chromate-reducing bacteria. The optimum pH for Cr(VI) reduction in both *Ochrobactrum* sp. CSCr-3 and *Pannonibacter phragmitetus* LSSE-09 was reported as 9.0 (He et al. 2009; Xu et al. 2011).

Cr(VI)-reduction by the G161 strain increased as cell inoculum volume increased from 1 % to 20 %, suggesting that Cr(VI) reduction is related to bacterial cell growth as observed in other species, such as *Bacillus* sp. (Kathiravan et al. 2011). However, higher inoculation volumes (over 30 %) failed to elicit further increases in Cr(VI) reduction,

suggesting either that bacterial concentration is rapidly saturated at 20 % of inoculum or that competition for limited nutritional resources restricts the bacterial metabolic activity in Cr(VI) reduction.

Cu^{2+} as a stimulator of Cr(VI) reduction was first reported in cell-free extract of *Bacillus* sp. ES 29 (Camargo et al. 2003b) and has been noted several times since (Sultan and Hasnain 2007; He et al. 2009; Xu et al. 2012). Again, our results are consistent with these previous investigations. By contrast, Ohtake et al. (1990) reported that the membrane-associated chromate reductase of *E. cloacae* was inhibited by 0.5 mM Cu^{2+} under anaerobic conditions. Addition of 50 mg/l Ni^{2+} and Zn^{2+} exerted no appreciable effect on Cr(VI) reduction by the G161 strain in our study, but other investigators reported that Zn^{2+} inhibits Cr(VI) reduction by *Ochrobactrum* sp. strain CSCr-3 (He et al. 2009) and *O. intermedium* SDCr-5 (Sultan and Hasnain 2007), while Ni^{2+} enhances Cr(VI) reduction in both strains. The Cr(VI) reductase activity of *P. phragmitetus* LSSE-09 is inhibited by Mn^{2+} under aerobic and anaerobic growth conditions (Xu et al. 2012), but this ion exerted no effect on Cr(VI) reduction in our present observations. Our results suggest that G161 is tolerant to a wide range of metal ions, although some, notably Ag^+ and Ba^{2+}, inhibit Cr(VI) reduction.

Wang et al. (2010) reported that Cr(VI) in solutions could be reduced to Cr(III) by indigenous bacteria under anaerobic conditions, at rates dependent on temperature and pH. In G161, the most effective wastewater Cr(VI) reduction was achieved under aerobic growth followed by facultative anaerobic incubation, consistent with our laboratory tests in in LB medium. This study suggests an important potential role for the isolated strain G161 in bio-treatment of chromate (VI) polluted wastewaters.

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