Assessment Of Cr (VI) Resistant Bacterial Diversity And Characterization Of Potent Chromium Reducers From Gwalior, India

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Abstract: Natural water resources are at high risk of heavy metal contamination by the introduction of untreated industrial, domestic and sewage effluents, that leads to scarcity of pure water. Access to potable water is a fundamental right of mankind as well as all living beings. This has led a high pressure on finding out most effective, economic and environment friendly approaches for sustainable management of natural water resources. Heavy metals are one of the major inorganic pollutants, highly toxic, irretrievable and affect the microbial community in polluted areas. Bioremediation is the most effective sustainable approach for heavy metal remediation. Since, there is a great microbial diversity in the environment; the isolation of indigenous Cr(VI) resistant bacteria has become a proven strategy for in situ bioremediation purposes. The present research work emphasizes on assessment of Cr(VI) resistant bacterial diversity in samples from different industrial sites and sewage outlets, Gwalior, Madhya Pradesh, India. A total of 69 chromium resistant bacteria were isolated from 20 different soil, sewage and effluent samples. Out of 69 bacterial isolates, 40% has showed Maximum Tolerance Level up to 25mg/L, 29% up to 50mg/L, 21% up to 75mg/L and 10% up to 100mg/L of Cr(VI) concentration respectively. On the basis of morphological and biochemical characterization, Cr(VI) resistant bacterial isolates with Maximum Tolerance Level up to 100mg/L were assigned to five genera- Bacillus (64%), Enterococcus (12%), Micrococcus (12%), Brevibacterium (6%) and Corynebacterium (6%). During Cr(VI) reduction study, 47% bacterial isolates have showed above 98% reduction and 26% bacterial isolates showed above 50% reduction at 25mg/L Cr(VI) concentration at 37ºC, 120 rpm up to 48h of incubation period. So, the present study could be helpful in assessing microbial diversity of Cr(VI) resistant bacteria from Gwalior and efficient chromium reducing isolates could be explored and identified for further bioremediation study.

Index Terms: Bioremediation, Effluent, Hexavalent chromium, Microbial diversity, Sustainable.

1 INTRODUCTION

THE continuous emission of heavy metals in the environment due to unprecedented industrialization and non-judicious use of resources has become a serious threat for ecosystem and human health. Heavy metals are natural constituents of earth and are essential in low concentration but at high concentration, they exert severe harmful impacts on environment and mankind. These are highly toxic because of their persistent nature and widespread research on advanced techniques for rectifying the toxic metals and other pollutants is going on [1]. Cr(VI) is one of the toxic and a well-known group- A human carcinogen due to its nature of carcinogenicity [2], teratogenicity [3], mutagenicity [4] in humans, animals and plants [5], [6]. Acceptable limit of total chromium content in drinking water is 50µg/L according to World Health Organization [7].

Gastrointestinal bleeding, tuberculosis and asthma are common ailments observed among the workers. Infertility, birth defects and stillbirths have also been recorded [8]. In a recent research work by Sharma and Gupta in 2018a, Lactic acid bacteria showed a good resistance against different heavy metals and their use as probiotics was also evaluated. So, these bacteria could be used as probiotics as well as in detoxification of toxic heavy metals from human body [9]. Indigenous microbes are efficient in bioremediation and useful in cleanup of polluted environments [10]. Microbial diversity has gained much interest in bioremediation due to its adaptability and the search for identification of new capable strains continues, as least number of microbial species in the environment has been explored. A number of chromium resistant bacteria have been isolated from tannery effluent [11], [12], river sediment [13], industrial sludge [14], [15], electroplating effluent [16], activated sludge [17], evaporation ponds [18] and Sewage and effluent [19]. For isolation of novel microorganisms and exploring their properties, cultivation-dependent research work is significant [20]. The present research study was aimed to explore the Cr(VI) reducing bacterial diversity from different industrial sites- Birla Nagar, Malanpur, Banmore and sewage sites in Gwalior, Madhya Pradesh, India. This will widen our knowledge of dominant chromium reducing bacterial genera in soil and effluent samples of Gwalior. Considering the significant environmental applications of potent Cr(VI) reducing bacterial isolates in detoxification of hazardous toxic Cr(VI) from contaminated sites in in situ applications, screening of potent chromium resistant bacterial isolates, their characterization and chromium reducing efficiency was evaluated for further possible utilization in bioremediation.

2 METHODOLOGY

2.1 Sample collection and processing

A total of 20 samples were collected from different industrial sites including soil, effluent and sewage samples, Gwalior, India. Samples were transported to laboratory at 4ºC by using cooling ice packs and analyzed for- pH and temperature by using standard methods.

2.2 Enrichment, growth conditions and isolation of Cr(VI) resistant bacterial isolates

Isolation of chromium resistant bacteria was done by serial dilution method on nutrient agar plates amended with 10mg/L Cr(VI) concentration at 37°C and120 rpm for 24h using shaker

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incubator (REMI CIS- 24 PLUS) after enrichment of samples by modified method by Joutey et al., 2011[21]. Enrichment of samples was done by adding 1g soil/1mL sewage or effluent in 50mL nutrient broth having 25mg/L of initial Cr(VI) concentration. After 24h of incubation, 1ml suspension from previous enrichment was transferred in nutrient broth with gradually increased Cr(VI) concentrations i.e. 50mg/L and 100mg/L respectively. The phenotypically diverse bacteria were isolated on nutrient agar by serial dilution method and spread plate technique. Chromium resistant isolates in pure culture form were maintained on nutrient agar slants at 4°C for further study.

2.3 Chromium tolerance (Maximum Tolerance Level) of bacterial isolates

Chromium resistant isolates were screened out for Maximum Tolerance Level against Cr(VI) by visual comparison method given by Malik and Jaiswal, 2000 [22]. Isolates were aseptically streaked on nutrient agar plates having successive Cr(VI) concentrations- 25, 50, 75 and 100 mg/L and incubated at 37°C for 24-48h. The highest concentration supporting bacterial growth was taken as Maximum Tolerance Level for individual isolates.

2.4 Morphological and biochemical characterization of potent Cr(VI) resistant bacterial isolates

Bacterial isolates having Maximum Tolerance were characterized morphologically and biochemically according to Bergy's manual of determinative bacteriology by Holt et al., 1994 for assigning their taxonomic identity [23].

2.5 Optimization of growth conditions for Cr(VI) reduction

Optimization of different growth conditions like- pH, temperature, initial Cr(VI) concentration and initial inoculum level for Cr(Vi) reduction study by bacterial isolates was performed. Abiotic controls were also used for monitoring abiotic reduction for each isolate. Broth suspensions were withdrawn regularly at 24h time intervals for monitoring growth by measuring optical density at 600nm and remaining Cr(VI) concentration was determined in culture filtrate (centrifuged at 10,000 rpm for 15 min in cooling centrifuge (REMI C-24 PLUS) by diphenylcarbazide method at 540 nm by Shimadzu UV-1800 spectrophotometer [24].

2.5.1 pH

The isolates were grown at different pH values (2, 4, 6, 8, 10 and 6.8) in Nutrient broth having 25mg/L Cr(VI) by using concentrated HCl and H_2SO_4 and incubated at 37ºC, 120 rpm for 72h.

2.5.2 Temperature

For evaluating effect of temperature on Cr(VI) reduction, isolates were inoculated in nutrient broth having 25mg/L Cr(VI) concentration and were incubated at different temperatures (20, 30, 40, 50 and 37ºC) and 120 rpm for 72h.

2.5.3. Initial inoculum level

For evaluating effect of the initial inoculum level, isolates were grown in nutrient broth having 25mg/L of Cr(VI) concentration by inoculating different amount of inoculum- 100 µl, 300 µl and 500 µl, at 37ºC, 120 rpm for 72h.

2.5.4 Initial Cr(VI) concentration

For evaluating effect of the initial Cr(VI) concentration, isolates were grown in nutrient broth having different Cr(VI) concentrations- 25mg/L, 50mg/L, 75mg/L, 100mg/L and 125mg/L at 37ºC, 120 rpm for 72h.

2.5.5 Chromium reduction efficiency of bacterial isolates

After optimization of different growth conditions, different bacterial isolates were evaluated for chromium reducing efficiency at optimum conditions of temperature, pH, inoculum and initial Cr(VI) concentration. Bacterial growth and remaining Cr(VI) concentration was monitored by withdrawing sample broth and culture filtrate at regular time intervals.

3 RESULTS-

3.1 Sample collection and processing

A total of 20 samples were collected from different sites, Gwalior including 14 soil samples, 3 sewage and 3 effluent samples. Details of collection sites, temperature, pH of samples and no. of obtained isolates is presented in Table-1. pH of the samples was in the range from acidic (pH 3.31) to slightly alkaline (pH 8.38) while, temperature varied in the range of 15-42ºC.

3.2 Isolation of chromium resistant bacterial isolates and determination of MTL

After enrichment of 20 different soil, sewage and effluent samples, number of cultivable chromium resistant bacteria was 69 (Table-2). Isolation was performed by serial dilution of enriched sample suspension after 72h of incubation at 37ºC on nutrient agar (10mg/L Cr(VI). On determination of Maximum Tolerance Level against different Cr(VI) concentrations, only 10% of bacteria have showed resistance up to 100mg/L of chromium concentration (Fig-1). Percentage of bacteria having MTL up to 50mg/L and 75 mg/L was 29% and 21%, respectively.

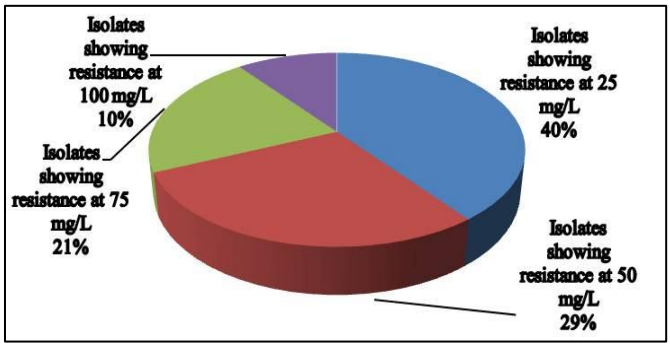

Fig.1. Percentage distribution of chromium resistant bacteria with different MTL.

3.3 Morphological and biochemical characterization of bacterial isolates

A number of 69 morphologically different aerobic bacteria were isolated from different samples, obtained in pure form and preserved at 4ºC for further experimental work (Table-2). All 18 chromium resistant isolates were Gram positive, while 72% isolates were motile and spore forming. On the basis of biochemical characterization, chromium resistant bacterial

isolates (18) showing maximum Cr(VI) resistance were tentatively assigned to 5 different genera- Bacillus, Enterococcus, Micrococcus, Brevibacterium and Corynebacterium, according to Bergy's manual of determinative bacteriology (Table- 3). As depicted in pie chart (Fig- 2), that the Bacillus (64%) was dominating among other chromium resistant bacterial genera. Enterococcus and Micrococcus were contributing 12% of total isolates, while Brevibacterium and Corynebacterium contributed 6%.

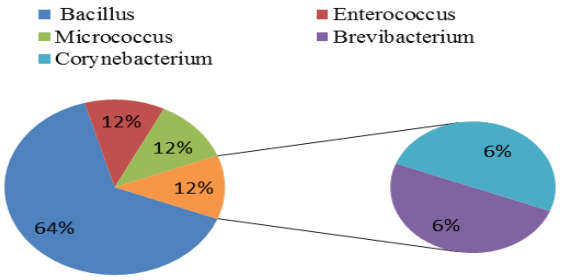

Fig.2. Percentage distribution of chromium resistant isolates.

3.4 Optimization of growth conditions

Evaluation of different crucial growth conditions including- pH, temperature, initial inoculum level and initial Cr(VI) concentration affecting chromium reduction by different isolates was done (Table-4). Optimum pH value was 6.8 or 8.0, temperature range was 30-42ºC and optimum inoculum size were 100 to 500 µl and optimum Initial Cr(VI) concentration was 25mg/L for all 18 bacterial isolates.

TABLE1 *SAMPLE CODES, COLLECTION SITES, TEMPERATURE, pH AND ISOLATES OBTAINED*

Sample codes	Sample collection site	Temperature	pH	Isolates Obtained
S.1.	Botany department, Jiwaji University, Gwalior	16°C	6.6	03
S.2.	Botany department (Wet), Gwalior	15°C	7.5	04
S.3.	Botany department (Dry), Gwalior	18°C	8.1	01
S.4.	Chetakpuri Gate Dumping Site, Gwalior	17°C	7.7	03
S.5.	Vivek Vihar dumping Site	22°C	7.2	04
S.6.	DRDO Dumping site	20°C	7.6	03
S.7.	AG Office Road dumping Site, Gwalior	22°C	7.2	03
S.8.	Govind Puri Dumping site, Gwalior	18°C	7.3	03
S.9.	Govind Puri Dumping site, Gwalior	19°C	7.0	03
S.10.	Sewage site Sharma form (Rayru road), Gwalior	35°C	7.4	05
S.11.	Battery manufacturing industry, Ganga Malanpur	40°C	4.75	1
S.12.	Gas Cylinder Plant, Banmore	42°C	7.75	06
S.13.	BCG Petrochemical Plant, Banmore	40°C	8.38	05
S.14.	IGNOT manufacturer of Steel, Banmore	42°C	8.08	05
SW.1.	Swarna Rekha nadi, Gwalior	19.1°C	7.6	04
SW.2.	Loco Road (NALA), Gwalior	18.9°C	7.4	03
SW.3.	Main Sewage Outlet of Gwalior, Sharma form, Rayru road	33.1°C	3.31	04
EF.1.	Loco car wash (Drain water), Gwalior	19.3ºC	7.6	03
EF.2.	Lead Industry, Ganga Malanpur	30.5ºC	7.17	03
EF.3.	Whey Plant, Banmore	30.1ºC	7.69	03

3.5 Chromium reduction efficiency of bacterial isolates

Chromium resistant bacterial isolates were evaluated for their chromium reducing ability at optimum values of growth conditions- pH, temperature, inoculum level and Cr(VI) concentration. During chromium reduction study by growth monitoring it was clear that maximum isolates gained exponential phase within 48h of incubation time (Fig-3) and later growth was either increased or remained static. Only S10C bacterial isolate showed low growth, while others showed well growth. Chromium reduction was evaluated by diphenylcarbazide method at 540nm, spectrophotometrically. Chromium reduction increased with increase in time of incubation up to 72h. Four isolates S2D, S11A, S14D and SW2C representing 11% of total isolates showed complete Cr(VI) reduction at 72h of incubation.

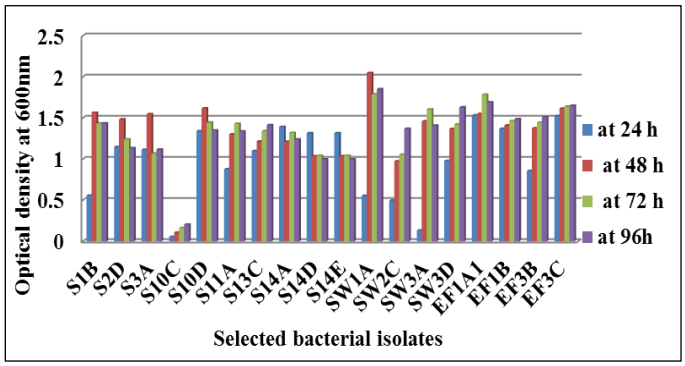

Fig.3. Growth of chromium resistant bacterial isolates at optimized growth conditions.

Percentage of chromium reducing isolates showing chromium reduction up to 75% was 23% (Fig- 4). Three isolates SW3A, SW3D, EF1A1 and S13C showed the minimum reduction.

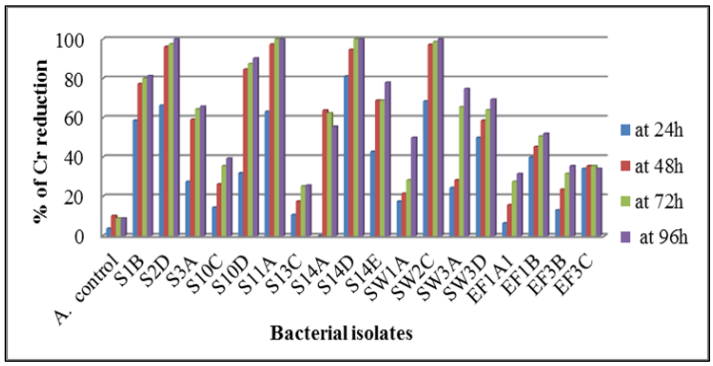

Fig.4. Percentage of Cr(VI) reduction (at 540nm) by chromium resistant isolates at optimized growth conditions.

***TABLE 2** MORPHOLOGICAL CHARACTERIZATION OF CR(VI) RESISTANT BACTERIAL ISOLATES*

S. NO.	Isolates	Colony characteristics	Gram's nature	Endospore	Motility
1.	SW1A	Cream, punctiform, entire, moist, raised	G+, rod	-	Motile
2	SW2C	Cream, small, entire, mucoid, transparent, raised	G+, short rod	-	Motile
3	SW3A	Colourless, punctiform, entire, moist, raised	G+, cocci	-	Motile
4	SW3D	Cream, small, entire, moist, opaque, raised	G+, short rod	present	Motile
5	EF1A1	Greenish, medium, wavy, moist and raised	G+, cocci	present	Motile
6	EF1B	Greenish, wavy, moist, transparent, raised	G+, diplococci	-	Motile
7	EF3B	White, punctiform, entire, moist opaque, raised	G+, rod	Central endospore	Motile
8	EF3C	Cream, small, moist, opaque, raised	G+, rod	Present	Motile
9	S1B	White, small, entire, glossy, opaque, convex	G+, Coccus	-	Motile
10	S2D	White, large, undulate, glossy, opaque, flat	G+, thick rod	Central endosore	Motile
11	S3A	Cream, medium, entire margin, moist, flat, transparent	G+, Rod, chain form	Present	Motile
12	S10C	Yellow, punctiform, entire, moist, opaque, raised	G+, Cocci, cluster	-	-
13	S10D	Off white, medium, fringes margin, glossy, mucoid, opaque, convex	G+, rod, chain form	Present	-
14	S11A	Cream, large, undulate, small, dry, opaque, flat	G+, rod	Endospore	-
15	S13C	Cream, medium, wavy, moist, opaque, flat	G+, short rod	Present	-
16	S14A	Cream, large, entire, moist, opaque, convex	G+, rod	Central endospore	-
17	S14D	Cream, medium, smooth, undulate, opaque, convex, glossy	G+, rod	Subterminal endospore	Motile
18	S14E	Off white, large, fringes, dry, translucent, flat	G+, rod	Central endospore	Motile

***TABLE 3** BIOCHEMICAL CHARACTERIZATION OF CR(VI) RESISTANT BACTERIAL ISOLATES*

Isolate Names	Sugar fermentation Test						IMVIC Test				Cat. Test	Oxi. Test	Gela. Test	Nitrate Red. Test	Ure. Test	Amy. Test	Tentative identification
	D	M	L	A	X	ML	I	MR	VP	SC							
SW1-A	+	+	-	-	-	-	-	+	+	-	+	+	-	+	+	+	Bacillus cereus
SW2-C	+	+	-	-	-	+	-	+	p+	+	+	-	-	+	+	-	Brevibacterium casei
SW3-A	+	P+	-	+	+	-	-	+	p+	-	-	+	-	+	-	-	Enterococcus faecalis
SW3-D	+	-	-	+	+	+	-	+	+	+	+	+	-	+	+	-	Corynebacterium Xerosis
EF1-A1	+	+	-	-	-	-	-	+	+	-	+	-	-	+	+	-	Miicrococcus variens
EF1-B	+	P+	-	-	-	-	-	+	-	-	+	+	-	+	+	-	Micrococcus variens
EF3-B	+	+	-	-	-	-	-	+	-	-	+	-	-	+	+	+	Bacillus cereus
EF3-C	+	+	-	-	-	-	-	+	+	-	+	+	-	+	+	-	Bacillus cereus
S1-B	+	P+	-	+	+	+	-	+	+	-	-	-	-	-	-	-	Enterococcus faecalis
S2-D	+	+	-	+	-	+	-	+	-	-	+	+	-	+	+	+	Bacillus thuringiensis
S3A	+	+	-	+	-	-	-	+	-	-	+	+	-	+	+	+	Bacillus cereus
S10-C	+	+	+	+	+	+	-	+	+	-	+	-	-	+	-	-	Staphylococcus aureus
S10-D	+	+	-	+	-	+	-	+	-	-	+	+	-	+	+	+	Bacillus megaterium
S11-A	+	+	-	-	-	-	-	+	+	-	+	-	-	+	+	-	Bacillus subtilis
S13-C	+	+	-	+	+	-	-	+	+	-	+	+	-	+	+	+	Bacillus cereus
S14-A	+	+	-	-	-	-	-	+	+	-	+	-	-	+	+	-	Bacillus subtilis
S14-D	+	+	-	+	-	+	-	+	p+	-	+	+	-	+	+	-	Bacillus cereus
S14-E	+	-	-	-	-	-	-	+	p+	-	+	+	-	+	+	+	Bacillus licheniformis

(+)- Positive, (-)- Negative, P+- partial positive, D- Dextrose, M- Maltose, L- Lactose, A- Arabinose, X- Xylose, ML- Mannitol, I-Indole, MR- Methyl Red, VP- Voges proskauer

***TABLE 4:** OPTIMUM VALUES OF DIFFERENT GROWTH CONDITIONS FOR CHROMIUM RESISTANT BACTERIAL ISOLATES*

Isolate names	Optimum pH	Optimum Temperature (ºC)	Optimum Inoculum level (µl)	Optimum initial concentration (mg/L)
SW1A	8.0	30	100	25
SW2C	8.0	37	500	25
SW3A	6.8	40	100	25
SW3D	6.8	37	100	25
EF1A1	8.0	37	100	25
EF1B	6.8	40	100	25
EF3B	8.0	40	100	25
EF3C	8.0	40	300	25
S1B	8.0	37	100	25
S2D	8.0	37	300	25
S3A	8.0	37	100	25
S10C	8.0	37	100	25
S10D	8.0	37	100	25
S11A	6.8	37	300	25
S13C	8.0	37	100	25

S14A	6.8	37	500	25
S14D	6.8	37	500	25
S14E	6.8	37	100	25

4 DISCUSSION

Chromium reducing bacteria have been isolated from different environments and research work is still in bottle neck development as the percentage of cultivable bacteria in laboratory conditions is very low and most of other novel bacteria are uncultivable. In the present study, the percentage of chromium resistant bacterial isolates belonged to genera Bacillus was high and other genera were- Enterococcus, Micrococcus, Brevibacterium and Corynebacterium at low percentage. Similarly, a high proportion of Cr(VI) resistant bacteria isolated from chromate polluted soil [25] and from tannery effluent [26] belonged to genus Bacillus. Significant Cr reduction by above mentioned bacterial sp. already has been reported by- Enterococcus [27], Micrococcus [28] [29], Brevibacterium [30] [31] and Corynebacterium [32]. The first report on Cr reduction by Enterococcus gallinarum strain was reported by Sayel et al., in 2012, from tannery waste contaminated soil [33]. Similarly, Teles et al., (2018) reported the ten chromium resistant genera, isolated from Manus-Amazon- Acidovorax sp., Acinetobacter sp., Alicycliphilus sp., Bacillus sp., Comamonas sp., Enterobacter sp., Micrococcus sp., Proteus sp., Serratia sp. and Vagococcus sp. [34]. So, there is a long list of Cr resistant bacterial sp. and many more are to reported. He et al., 2016, had also reported isolation of Bacillus odysseyi YH2 species from chromium-contaminated waste water along with Zobellella denitrificans YH1, Pseudomonas stutzeri YH3, Lysinibacillus sphaericus YH4, Exiguobacterium profundum YH5 and Pseudomonas mendocina YH6. No prior reports are available on chromium reducing Zobellella species [35]. The number of Cr(VI) resistant bacteria was higher in soil samples than sewage and effluent. Kabir et al., 2018, had also reported that a greater number of Cr(VI) resistant bacteria were obtained from solid wastes while a lower one from effluents. This may be due to regular deposition of Cr(VI) in soil. Sewage and effluent samples had a number of salts and other toxicants that inhibit bacterial growth [36]. Cr(VI) reduction is affected by a number of growth conditions like pH, temperature, inoculum level and Cr(VI) concentration, it directly affects the growth of resistant bacteria, therefore reduction also decelerates down. Optimization of growth conditions was done to achieve complete Cr(VI) reduction by bacterial isolates. In the present Cr(VI) reduction study, most of bacterial isolates showed maximum reduction at neutral pH 6.8 and for others optimum value was pH 8.0. Temperature optimization has revealed that optimum temperature was 37ºC for most of isolates and for only four isolates was 40ºC. In general optimum pH and temperature for bacterial growth was pH 6.8 and 37ºC. Kabir et al., 2018, has also reported that all the bacterial isolates grew and reduced Cr(VI) at a wide range of temperatures ranging from 25 to 45°C. The growth and Cr(VI) reduction by all the bacterial isolates increased with increase in temperatures up to 40°C and decreased above 40°C. In another study by Rehman and Faisal (2015) reported that the optimum pH and temperature for Cr(VI) reduction by Bacillus pumilis at 200 ug/mL Cr concentration was 7.0 and 37ºC, respectively [37]. Inoculum level is also a crucial factor for Cr reduction as the nutritional competition and variability in survival mechanism in presence of toxic Cr concentration may be the reason. In the present study optimum inoculum level for most of isolates was 100 µl and for others was also 300 and 500 µl. Effect of initial Cr(VI) concentration on Cr reduction was also evaluated and all the isolates have showed maximum reduction at optimum initial Cr(VI) concentration of 25mg/L. All isolates showed well growth up to 50mg/L but with increasing concentration there was a sharp decrease in growth. Some isolates also showed growth up to 100mg/L of Cr(VI). Similar results were also reported by Degiam [38] and Ramesh [39]. However no correlation was observed between Cr(VI) resistance and reduction [40]. Similarly in our study there was no relation between resistance and reduction of Cr(VI) by some bacterial isolates as SW3D and EF1A1 showed good resistance at 25mg/L Cr(VI) concentration but reduction was very low, while SW2C had a poor growth but reduced Cr(VI) within 72h of incubation completely. Bacillus subtilis has showed 9% of Cr(VI) reduction at 100mg/L and 5% at 50mg/L of initial Cr concentration respectively [41] fter optimization Cr(VI) reducing efficiency of bacterial isolates was evaluated by inoculating in nutrient broth at optimum conditions of inoculum, pH, temperature (37º/40ºC) and 25mg/L Cr(VI). In the present study 27% of isolates showed ≥40-60% of chromium reduction at 25mg/L Cr(VI). Only 4 isolates (SW2C- Brevibacterium casei, S2D- Bacillus thuringiensis, S11A- Bacillus subtilis and S14D- Bacillus cereus) representing 11% of total isolates showed complete chromium reduction. S1B- Enterococcus faecalis and S10D- Bacillus megaterium also showed excellent Cr(VI) reduction of 80% and 90%, respectively. In another study by Haddar et al., 2018, B. megaterium A3-1 has showed complete removal (100%) of Cr at 30 mg/L after 72h at 37°C [42]. According to Sen et al., 2018, CRB 33 isolate also showed 93.4% of Cr reduction at 100mg/L after 24h [43].

5 CONCLUSION

Present study revealed that there is a wide diversity of Cr(VI) resistant bacteria in different samples from Gwalior, Madhya Pradesh. A total of 18 chromium resistant bacteria has been isolated and taxonomically identified to belong five genera- Bacillus, Enterococcus, Micrococcus, Coryneybacterium and Brevibacterium. On evaluation of chromium reduction efficiency of isolates, Bacillus species have showed excellent chromium reducing ability. Exploration of different environments for isolation of heavy metal reducing bacteria would enhance the databases for diversity of some novel resistant bacteria. These bacterial isolates in the present study could be further assed for utilization in bioremediation approaches of toxic heavy metal detoxification in a sustainable manner.

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CONFLICT OF INTREST

There is no conflict of interest among authors on any aspect.

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