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Heavy metals pollution in iron tailing soil and its effects on microbial communities metabolism and enzyme activities

The purpose of this research is to examine the relationship between the microbial communities and heavy metals, the activities of enzymes in tailing soils of iron ore in the upstream area of Hanjiang river in Shaanxi, China was a case study. Six soil samples were gathered from the tailing region at different distances. The samples were then grouped into two parts, one was sieved with a nylon sieve of 0.149μm, in other to obtain its chemical composition, and the type of heavy metals present, and the other was analyzed using the CLPP and soil enzymes test after been stored at 4ºC. Metals were digested and measured in ICP-AES solutions. Biolog ECO-micro plates was used to determine the community-level physiological profiles (CLPP).

Results and discussions of tailing soils of iron ore shown a significant variation, mainly manifested in the metabolic patterns of carbon sources such as polymer and amino acids, with an increasing soil heavy metals pollution. The utilization of carbon sources by microbial communities was activated in slightly polluted soils and was constrained in heavily polluted soils. Activities of soil urease, alkaline phosphatase, catalase and dehydrogenase declined with intensified soil heavy metals pollutions. The soil urease, protease, alkaline phosphatase and catalase activities in the tailing center site were 9.91%~21.19%, 29.94%~37.16%, 24.15%~29.90% and 10.0% lower than those in the farthest distance from the center respectively. The soil content affected; urease, alkaline phosphatase, and catalase which were negatively correlated.

Key words: *Heavy metals pollution, soil microbial communities, principal component analysis, enzyme activity.*

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1. Introduction

The areas contaminated by heavy metals have expanded enormously during the last century, which is mainly due to urbanization and several industrial activities which include: mining, smelting and manufacturing. Currently, heavy-metal toxicity and accumulation have brought serious ecological problems. The heavy metals can mix with water that is leached into the soil and then absorbed by plants before being released into the atmosphere as gas or it could be bounded into soil components, such as clay or organic matter [1], seriously affecting human health [2, 3]

Mining, embodied smelting and metal processing activities, led to perturbations in cycling of metals in the surface environment [4]. For example, iron tailing causes heavy metal contamination and it also occupies a large expanse of land [5]. The biological accumulation that resulted from the transfer and migration of heavy metals in tailing areas, was observed, this also resulted to some changes in the soil properties. Furthermore, heavy metals posed severe treats to human health through food chain [6]. Most of the recent studies laid emphasis on the heavy metal pollution in non-ferrous metal mine (Cu or Pb-Zn mine) [7, 8, 9], while few other research paid attention to ferrous metal mine (Fe mine), especially deuterogenic environment effect of oxidizing iron mine tailings. Iron-ore mines have shown to be an important source of major metals, mainly Fe and Mn, also associated with traces of metals from the environment [10]. Research has often shown that there is a high level of heavy metals in iron tailing, which results to changes in the water and soil chemistry [11] and the bio-availability of metals. Xu Zheng-qi[12], Yang Jin-yan[13] analyzed the environmental impact caused by mining V-Ti-Mn in Panzhihua. Gao yan-xin [14], Huang Xing-xin [15] analyzed the soil heavy metal pollution around the iron mine exploration area in upper basin of Beijing Miyun reservoir. Xing Yi [16] studied the influence to the change of soil microbial community of heavy metal pollution in iron mining. The study generally suggests that the oxidizing mine already has significant influence (affected) on environment.

Microbes are more sensitive to heavy metals than animals and plants in soil. The changes of microbial activity and community structure could reveal the soil quality and health sensitively [17], thus soil microbes were considered the most efficient way of evaluating soil quality [18]. Some enzyme activities, such as inverts and peroxides, can be used in determining the changes of soil condition during spruce forest restoration [19]. A reassessment showed that excluding the study by Xing which showed the effects of heavy metal pollution on soil microbial community change in iron mining area using fluorescent PCR, few other researchers investigated the soil microbial metabolic activity, microbial community diversity and enzyme activity in the iron mining area.

The purpose of this work was to (1) study the level of heavy metal pollution in iron tailing, in Hanjiang upstream; (2) study the microbial community function in soil and obtain the relationship between enzyme activity on the soil and the heavy metal present in the soil; (3) explore the major factors that strongly influence the microbial communities in the iron tailing soils.

2. Materials and method

2.1. Description of study site

Hanjiang upstream is located at the mid-reach of "South-to-North Water Diversion Project" and the quality of soil and water there significantly affected the worth of this national project. At the time of smelting and mining of iron in Hanjiang upstream, waste backfilling was adopted there and the influence this had on the soil and water at the mining site was not studied. Mi Jian-gou tailing in Lveyang county (33°12'48.5"N, 106°24'52.5"E) in southwest of Shaanxi province, in Hanjiang upstream, China, was chosen as the sourcing area. Lveyang county lies in the south piedmont of Qin-Ling Mountain which has a warm temperate zone with moist monsoon climate. The annual average temperature is 13.2 ºC and annual average rainfall is about 860 mm. Lveyang, which is rich in iron mine, experienced poorly planned and wasteful mining of iron in the 1980s and 1990s. A lot of small and medium-sized tailings took natural valley as dam. Without a manual scale of reclamation of the slope and dam surface of these tailings, dust and rain infiltration could transport the heavy metals into nearby environment. The Mi Jian-gou tailing area is a rank-2 tailing and is still in use after 30 years.

2.2. Soil sampling

On October, 2014, we marked the center of tailings area as M2 sampling site and then determined other sampling sites considering the local wind (northwest wind) and their distance to M2. M1 was in 2 km upwind to M2 in wetland. In the downwind area, 4 sampling sites were marked as M3, M4, M5, and M6 which were selected according to their distance to M2 and vegetation. Soil samples were collected at a depth of (0~20cm) by random sampling. A soil sample consisting of soils collected from 3 points within a radius of 5 meters around the center site was obtained; this was done at every sample site. The samples were transported in a refrigerator to the lab, were it was divided into two parts. The first part was air-dried to determine the chemical properties and heavy metal contents, and then crushed, homogenized and sieved through 0.149μm nylon sieve. All prepared samples were stored in plastic containers for use. The second part was stored at 4ºC before the analysis of the CLPP (on the next day) and soil enzymes (within four days).

2.3. Measurement of soil chemical properties and heavy metal concentrations

Total nitrogen (TN) was estimated by Kjeldah's method (1883) and total phosphorus (TP) was estimated by that of Homer (1961). Total potassium (K) content was measured by flame photometer method (AA-6800, Japan). Organic matter (OM) was assayed by the dichromate oxidation method (Lu, 2000). The soil pH was measured in a suspension of 1: 5 soils to de-ionized water (Sharma et al. 2007) using pH meter (Starter-3C, OHAUS, USA) after 0.5 h agitation. The concentration of heavy metal was measured by ICP-AES (SP8000, Beijing Rayleigh Analytical Instrument Corp, China Beijing) after digestion. Blanks and replicates were adopted for quality control (Duzgoren-Aydin et al. 2006). The soil unease activity, catalase activity, alkaline phosphates activity, dehydrogenate activity were all determined using the method by Guan (1986).

2.4. CLPP

The destructive metabolic diversities of the microbial communities were estimated by Biolog ECO-micro plates (Biolog Co., Hayward, CA, USA) which consisted of 3 replications containing 31 carbon (C) sources and a control well without C source. 3 replicates of 5g fresh soil samples were dissolved in 45mL sterile NaCl solution (0.85%) and shaken for 30min at 200 rpm (revolution per meter). The suspension was step-wisely diluted to 10–3 using sterile NaCl solution (0.85%). Then 150uL diluted suspensions were inoculated to each well of the ECO-micro plate and incubated at 28ºC for 240 h. The utilization was measured with a Biolog micro station TM (BIO-TEK Instruments Inc., Winooski, VT, USA) at 590 nm for every 24 h.

2.5. Statistical analyses

All statistical analyses were carried out using Microsoft Excel 2003 and SPSS 19.0. Kinetic analysis was undertaken with average well colour development (AWCD) for Biolog to display the microbial activity: AWCD= (Ci–R)/31, in which Ci was the absorbance of 31C sources, R was the absorbance of the control well. The Shannon richness index (H), Simpson dominance index (1/D) and McIntosh all once index (U) were calculated using the data at 120 h, and the calculations were performed as described by Zhen Li-sha [20].

3. Results and discussion

3.1. Soil characteristics

Chemical properties of both the soil and the heavy metals are important indexes of their potential mobility in soil [21]. The pH, TN, TP, TK and organic material (OM) values of soils in the study area were collected as shown in Table 1. TN, TP, TK and OM increased with distance from the center of mine tailing marked as M2. The parameters in M1 and M3 all exceeded those in M2. Although M3 with reed grown was located in mine tailing area, its index of the chemical properties was higher than that of M1, especially OM which was 2 times that of M1. This indicated that vegetation cover could alter the tailing physical and chemical properties. M4, M5, M6 lied in the downwind of the tailing as agricultural land in which soil nutrient were significantly higher than those in M1, M2, M3 thanks to fertilization. The soil pH ranged from7.05 to 7.54 except for that in M4 (pH=6.91(SD±0.02)). No significant correlation between soil pH with the distance from M2 was found.

3.2. Heavy metals concentrations in the soils

Table 2 presented heavy metals concentrations of the soils, published reports of the background values as well as the secondary soil environmental quality standards from the Chinese Environmental Protection Administration [22]. As shown in Table 2, there was a decreasing value in heavy metals concentrations as the distance from M2 increased. In control site M1, Fe, Cu, V, Cd contents slightly exceeded the background values and other 6 elements were below the limit. While the metals concentrations of 5 other samples were all above background value. The exceptional ratio of heavy metal in M2 was highest in all 6 sites as a result of it being located at the heart of the iron tailing area. Among which, Cu was 15.32 times of background value. M4, M5 and M6 were all in agricultural land outside the tailing area. Heavy metal pollution in the sites closer to tailing center was more critical, suggesting that the tailing contributed enormously to the heavy pollution in the areas with the same direction of the wind. In M4, also the closest site to the tailing center, Ni, As, Cu, Hg and Cd contents were 3.01, 1.72, 3.07, and 1.72, which were about 5.13 times higher than the secondary soil environmental quality standards. In M5, which is located farther than M4 in proximity to the tailing center, , Fe, Mn, Zn and Ni contents were similar to those in M4, but As, Cu, Hg, Cd, V and Co contents were comparably different from M4 as a possible result of metals migration. In general, metal contents in M5 were lower than those in M4. In M5, Ni, Cu, Hg and Cd concentrations were 3.00, 1.07, 1.24, 2.47 times of

Table 1: The soil samples condition and mean PH, TN, TP, TK AND OM

Sampling sites	Distance from the tailing center (m)	Plants of sample	TN (%)	TP (%)	TK (%)	pH	OM (%)
M1	1000	Weeds (CK)	0.14±0.05	0.08±0.02	0.92±0.10	7.07±0.02	1.76±0.15
M2	0	None	0.09±0.01	0.07±0.01	0.78±0.09	7.31±0.05	0.71±0.17
M3	200	Reed (in tailing)	0.16±0.05	0.08±0.03	0.82±0.06	7.51±0.03	1.46±0.24
M4	600	Corn (out tailing)	0.15±0.04	0.10±0.02	1.25±0.21	6.91±0.02	4.84±0.36
M5	800	Pea (out tailing)	0.16±0.06	0.15±0.02	1.49±0.13	7.47±0.03	5.51±0.31
M6	1000	Rice (out tailing)	0.20±0.03	0.21±0.03	1.34±0.21	7.49±0.04	8.64±0.32

Notes: The values are the mean of three replicates (means ±SD)

Table 2: The statistics of heavy metal results ($mg.kg^{-1}$)

Heavy metals	Samples						Background values	Soil quality standard
	M1	M2	M3	M4	M5	M6		
Fe	43245	88868	70602	65476	61237	44157	28600	-
Mn	626	1304	1144	1192	1120	653	684	-
Zn	67.6	166	155	121	116	76.1	71.8	250
Ni	32.7	189	178	151	145	86.8	31.5	50
As	10.9	99.9	74.5	43.0	21.8	20.9	12.5	25
Cu	50.2	409	387	307	107	97.9	26.7	100
Hg	0.10	2.73	1.23	0.86	0.64	0.30	0.105	0.50
Cd	0.15	3.28	2.45	1.54	0.74	0.55	0.071	0.30
V	103	232	230	217	171	132	93	-
Co	17.8	92.6	61.6	56.7	32.6	29.8	16.2	-

Correlation analysis between heavy metal content, pH, organic matter (OM), TN, TP and TK

TABLE 3: CORRELATION OF HEAVY METAL CONTENTS, PH, OM (ORGANIC MATTER), TN, TP AND TK IN SOIL

Metals	pH	OM	N	P	K	Fe	Mn	Zn	Ni	As	Cu	Hg	Cd	V	Co
Fe	0.071	-0.568	-0.800	0.364	-0.456	1									
Mn	0.002	-0.402	-0.642	0.435	-0.180	0.925**	1								
Zn	0.206	-0.550	-0.745	0.344	-0.454	0.965**	0.918**	1							
Ni	0.257	-0.283	-0.528	0.204	-0.182	0.908*	0.844	0.954**	1						
As	0.156	-0.611	-0.791	0.287	-0.665	0.930**	0.760	0.926**	0.823*	1					
Cu	0.00	-0.543	-0.716	0.448	-0.575	0.893*	0.818*	0.924**	0.863*	0.944**	1				
Hg	0.111	-0.566	-0.796	0.219	-0.556	0.953**	0.744	0.879*	0.788	0.949**	0.837*	1			
Cd	0.132	-0.584	-0.779	0.313	-0.614	0.952**	0.809	0.948**	0.864*	0.996**	0.962**	0.946**	1		
V	0.054	-0.396	-0.612	0.406	-0.322	0.907*	0.937**	0.952**	0.963**	0.861*	0.946**	0.783	0.901*	1	
Co	0.023	-0.512	-0.748	0.303	-0.533	0.957**	0.828**	0.915*	0.855*	0.971**	0.942**	0.963**	0.981**	0.894*	1

Notes: *P<0. 05; **P<0. 01

those in the secondary soil environmental quality standards and the rest metals were within the standard concentration index. In M6, which is the farthest location to the tailing center, all metals excluding Ni and Cd were under the standard concentration. Ni and Cd exceeded the standard limits by 1.73 and 1.83 times, respectively. From the above, it was established that the soil around the tailing displayed pollution of heavy metals, such as Ni, As, Cu, Hg, Cd, V and Co, among which Ni and Cd were more critical.

In Table 3 presumed sources of heavy metals in soil and their controlling factors OM, TN, TK and the concentration of heavy metals display negative correlation without any sign of positive correlation, which implies that the chemical properties of the soil had negligible influence on heavy metal pollution. However, a significant positive correlation was shown between Fe and the 9 other metals. There was a high level of positive correlation between Fe and Mn, Zn, As, Cu, Hg, Cd and Co ($P<0.01$), which indicated that these metals had their source from an iron mine and Fe in soil particles could adsorb other metallic particles to form insoluble compounds (Forster,1989). Significantly positive correlation between Mn and Zn, V, Co in the sample soil (RZn=0.918, RV=0.937, RCo=0.828, $P<0.01$) were revealed, perhaps mainly due to transformation characteristics and migration of Mn. Significantly positive correlation between Zn, Cu, Cd, as and Ni ($P<0.01$) showed that these metals had same artificial and natural source.

3.3. CLPP

3.3.1. AWCD

The AWCD directly reflects the overall C source metabolic activity of microbes [23]. As shown in Fig.1, there were obvious differences among all treatments at 72h, although the AWCDs of M1 and M2 were similar after incubation for 96h. Significant difference in AWCD among all treatments was presented at 120 h. The overall AWCDs followed as: M6 > M4 > M5 > M3 >M1>M2; however, it should be pointed out that AWCDs of M5 was higher than that of M4 from 72h to 120h. The AWCD of M2 was significantly lower than that of other treatments at all time, reaching 0.55 at 240 h. 27.8% of that in M6. M2, which has the most critical pollution site, presented the greatest metabolic restrain for soil microbial communities. The heavy metals content in M4 were slightly higher than that in M5, but the metabolic activities of the soil microbial community of M4 was greater than that of M5, this indicates that these heavy metals promoted soil microbial communities. In general, the heavy metal contents were lower and the metabolisms of microbial communities were weaker when the sites were farther in proximity to the tailing center.

3.3.2. PCA

Heavy metal pollution not only affects soil microbial activity and changes the microbial community structure, but also introduces variability in the microbial metabolism and changes the selection and use of carbon source. PC1 and PC2 were determined by the PCA at 120h, and they contributed in accounting for 67.08% of the total variance, 58.18% and 8.90%, respectively. The factor loadings of the 31 C sources categories PC1 and PC2 were shown in Fig.2. As shown, PC1 mainly consisted of 14 components(eigenvector>0.80) including 4 amino acids, 3 polymerizations, 2 carbohydrates, 2 amines, 1carboxylic acid, which indicated that amino acids were the main differentiation of carbon source on PC1, followed by polymer and sugar. Whereas A2 (B-methy-d-glucosamin), E2 (n-acetyl-d-glucosamin) and G4 (a-ketobutyric acid) made major contribution to PC2.

The PC1 and PC2 scores are shown in Fig.3. The treatments were divided

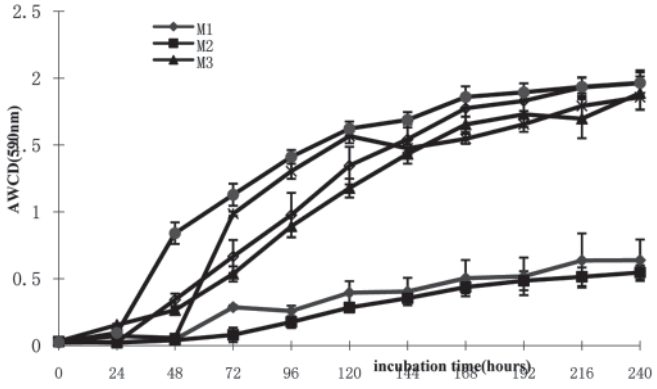

Fig.1 AWCD of soil microbial community during the incubation

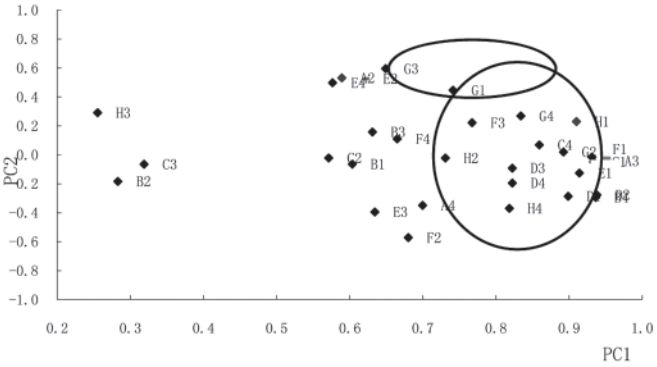

Fig.2 Loadings of 31 carbon sources on PC1 and PC2 in the principal components analysis of the CLPP

into 4 groups based on the PCA scores; as a result, M1 and M5, M3, M2, M4 and M6 were in 4 quadrants, respectively. The difference which was obvious on the distribution of the principal component in the coordinate system for the treatments was mainly manifested on PC1. M5 with the value of 1.08 had the highest score on PC1 while M2 and M3 (-1.42) had negative scores on PC1. The variance analysis showed that the values of typical PC1 variable were significantly different ($F = 179.21$, $p < 0.01$). The differences were extremely significant between M2, M3, M4 and M5. From the perspective of the discreteness of typical values of variables, there was less discrete (variation) of M1, M5 and M6, whereas more discrete of M2 and M3, suggesting the microbial community structure of soil in M1, M5 and M6 which were far from the tailing center were more relatively stable than the structures in M3 and M4 which were closer to the tailing center. On PC2, the differences also were extremely significant between M2, M3 and M1, M4, M6 ($F = 10.95$, $p<0.01$).

3.3.3. Carbon source metabolic analysis of microbial community

Differences in the source of carbon can reflect the metabolic function in soil microbial community. By Biolog method, in combination with the utilization of different carbon source in soil microbial community analysis at 120h, it was

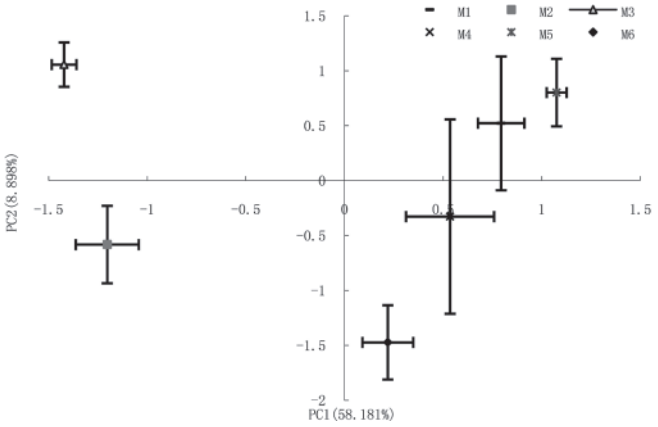

Fig.3 Principal component analysis of soil microbial community

found that the priority kind for the use of carbon source and utilization degree obviously varied in different soil microbial community (Fig.4). All the sample soils could utilize 6 substrate categories and the utilization levels varied from 0.29 to 1.96. M2 had the lowest utilization levels of all 6 substrate categories in all sites. The utilization levels of 6 groups of carbon source in M2 and M3 were significantly lower than that in other sites. M5 had the highest utilization of amino acids, polymers and carbohydrates in the samples. M6 presented a relative higher utilization of carboxylic acids, aromatic compounds, and amines. In all sites, the utilization levels of carbohydrates and aromatic compound were the lowest.

Microbial community diversity index like Shannon richness index (H), Simpson dominance index (1/D) and McIntosh all once index (U) were often used to evaluate soil microbial functional diversity [24]. To be specific, H reflected the richness of soil microbial community; 1/D focused on the most common species in soil microbial community and U was based on the species diversity index of the multidimensional space distance which was the community species homogeneity measurement. Table 4 showed the diversity index calculated using the Biology data at 120h. H indicated that the soil in M1, M4, M5 and M6 were significantly ($P < 0.05$) higher than those in M2 and M3. Notably, M2 had the lowest values of H, S and U, and M5 had the highest corresponding values. There were no significant differences in the values of H that were detected between M1, M4, M5 and M6. S did not differ significantly among M1, M3, M4, M5 and M6, however, S in all other treatments was significantly higher than that of M2.

3.4. Enzyme activity

Table 5 shows that enzyme activities had a similar trend. With distance from M2 increasing, the pollution reduced and activity increased. There was a significant difference in urease, catalase and dehydrogenase. Enzyme activity in M2 were 9.91%?21.19%, 29.94%?37.16%,24.15%?29.90% and 10.0% lower for urease, catalase, alkaline phosphatase and

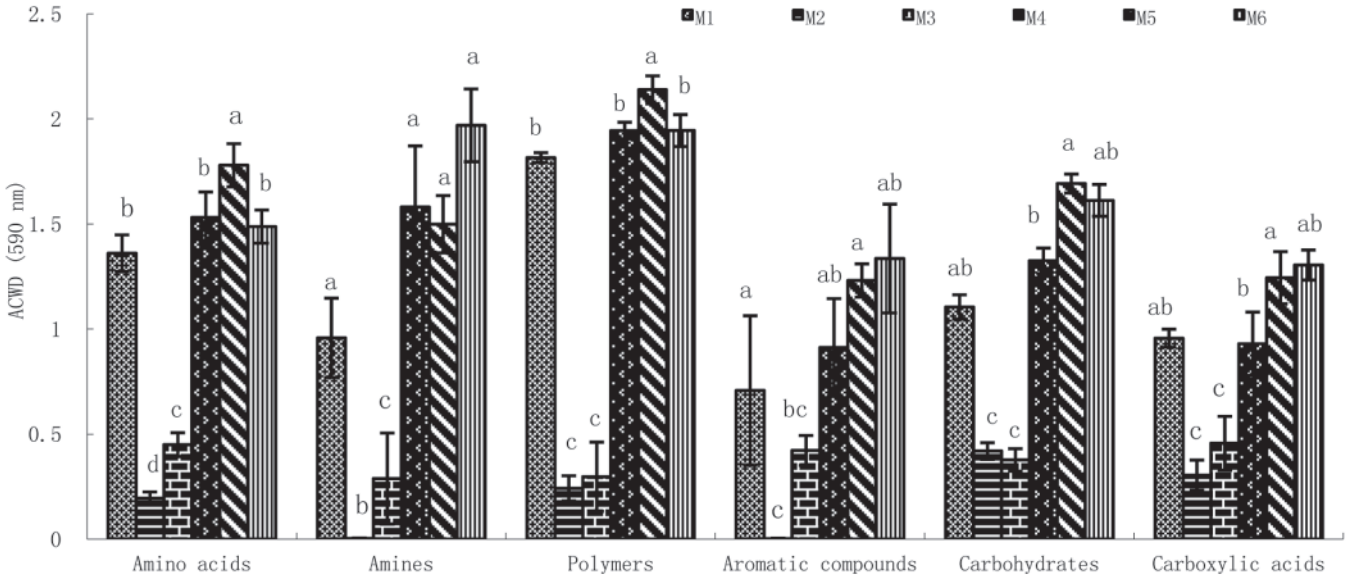

Fig.4 Utilization of carbon source by soil microbial community

TABLE 4: THE DIVERSITY INDICES OF SOIL MICROBIAL COMMUNITY

Samples	Shannon (H)*	Simpson (1/D)*	McIntosh (U)*
M1	3.297±0.011 a	46.474±3.906 a	8.969±0.021b
M2	2.289±0.240 c	35.175±3.807b	3.249±0.531c
M3	2.871±0.213 b	47.528±5.874 a	3.344±0.529c
M4	3.235±0.028a	50.157±3.232a	8.005±0.453b
M5	3.298±0.016a	51.302±0.110a	10.014±0.202a
M6	3.139±0.028a	49.704±1.842a	9.673±0.168 a

Notes: The values are the mean of three replicates (means ± SD). *Values followed by letters differs significantly (LSD test, P<0.05)

TABLE 5: ACTIVITIES OF THE SOIL ENZYMES

Samples	Ure*	Alkali-pho*	Cat*	Dehydrogenase*
M1	11.02±1.57c	102.83±14.98b	25.65±1.90d	0.35±0.05c
M2	2.10±0.90d	63.61±14.98c	14.25±1.90f	0.15±0.01d
M3	3.67±0.91de	76.68±0.01bc	18.68±1.45e	0.47±0.05c
M4	15.74±1.57b	181.28±39.63a	38.32±1.45c	1.54±0.15b
M5	19.94±2.40a	187.82±24.68a	43.07±1.45b	1.81±0.12a
M6	12.59±1.57c	191.09±14.98a	52.57±1.45a	1.50±0.10b

Ure: Urease (ug NH_3-N/g·d); Alkali-pho: Alkaline phosphatase (ug hydroxy-benzene/g·d); Catalase (mg/g·d); Dehydrogenase (g/g.d); *Different letters in the same column means significant difference at 0. 05 levels (P<0.05).

dehydrogenase, respectively than those in M6. The overall alkaline phosphatase and catalase activity presented in the following order: M6>M5>M4>M1>M3>M2, which are closely associated with the soil heavy metal pollution. The order of urease and dehydrogenase activities is as follows: M5>M4>M6> M1>M3>M2. M5 displayed the highest level of urease and dehydrogenase activities, which was significantly different from other sampling soils. It can be concluded that dehydrogenase and urease enzyme effected activation of lower pollution level on soil, and restrained higher levels of pollution. All the enzyme activity levels of M4, M5 and M6 were higher than those of M1 (CK), probably because M4, M5 and M6 were in crop land where organic fertilizer can increase the biomass and bioactivity, especially the nitrogen and phosphorus

TABLE 6: CORRELATION COEFFICIENTS OF SOIL ENZYME ACTIVITY AND HEAVY METAL CONTENT

Enzyme activity	Fe	Mn	Zn	Ni	As	Cu	Hg	Cd	V	Co
Urease	-0.542	--0.232	-0.560	-0.347	-0.793*	-0.671*	-0.677	-0.741*	-0.431	-0.661
Alkali-phosphatase	-0.509	-0.232	-0.515	-0.242	--0.672*	-0.518	-0.603	-0.618	-0.310	-0.517
Catalase	-0.642	-0.438	-0.633	-0.372	-0.729*	-0.646	-0.668	-0.698	-0.479	-0.623
Dehydrogenase	-0.369	-0.063	-0.340	-0.051	-0.568	-0.422	-0.506	-0.507	-0.163	-0.429

*P<0. 05

fertilizer applied made the urease and phosphatase activity increase significantly.

Correlation analysis (Table 6) showed that urease, alkaline-phosphatase, catalase and dehydrogenase activity were negatively correlated with heavy metal contents. The correlation between Cu, Cd content and urease, was found to be significantly negative ($P<0.05$). Similarly, there were highly significant negative correlation between As content and urease alkaline, phosphatase and catalase (P?0. 05). And no significant correlation between enzyme activity and other heavy metals was found.

4. Discussion

Mining has seriously brought with it an increasing soil erosion and environmental pollution caused by manufacturing waste in extraction beneficiation and smelting of minerals and high heavy metals content in the tailings polluted water and soils. Bioremediation was a potential microorganism-based remediation technique to degrade and detoxify heavy metals contaminants [25]. These biological systems are cost effective and can compensate for the disadvantages of less amenable activities to environmental extremes, than would by traditional methods [26]. Soil microorganisms were important components in maintaining soil biological activity because they were more sensitive to interference. As a result, microbes have been used as an important and indispensable biological indicator to evaluate soil quality. Teng Ying [27] concluded that soil microbial community functional diversities decreased in soils polluted by heavy metals such as Pb, Zn, Ag from mine tailing. Additionally, decreased microbial number used energy carbon and utilization ability of microbial communities for carbon substrates. In attempt to give a well detailed mode of interaction between heavy metal contamination and microbial diversity, Xie Xue-hui [28] studied the heavy metal pollution in copper mine tailing. The laboratory study showed different result compared to the study on aged wild field that were contaminated by heavy metals in which the relationship between heavy metals concentrations and microbial diversity is not a simple linear function. In the present study, it is shown that soil microbial community in farther sampling sites with slightly heavier metal pollution had activation effect on utilizing carbon source; however, in M2 where there was heavy metals pollution, the effect was restrained. Average colour change rate (AWCD) was the highest in moderately and mildly polluted sites while the lowest AWCD are presented in severely polluted sites. This may be as a result of difference in climate conditions and different types of heavy metal pollution in mine soil. In the mine areas of Pb, Zn, Ag and Cu, heavy metals were the main pollutants with higher contents than that in Fe mine area than it is in this study in which Cd and Ni were the main pollutants. Simultaneously, this study showed that mild and moderate pollution could improve the Shannon richness index (H), Simpson dominance index (1/D) and McIntosh index (U) of soil microbial community. There was an obvious inhibition effect on U, a certain inhibition effect on H and 1/D, but no obvious uniformity effect was observed in severely polluted sites. This was differentiated from coal mining area where there was obvious inhibition effect on H, 1/D and U of soil microbial community in severely polluted soils [29].

Soil enzyme activity reveals soil biological activities, the soil fertility characteristics and soil nutrient transformation process. The potential biological toxicity of pollutants was assayed from the sensitivity of soil enzyme to heavy metal pollution. Urease enzyme played important roles in soil nitrogen utilization and nitrogen cycle through catalyzing the decomposition reaction of ammonia urea in soil. Phosphatase activity is the index for evaluating the direction and strength of soil phosphorus biotransformation and may accelerate the dephosphorization of organic phosphorus. Catalase enzyme participates in the metabolism of biological respiration in which aerobic activity is closely linked with biomass and soil fertility. Dehydrogenase, a kind of intracellular enzyme, converses hydrogen transfer by catalyzing dehydrogenation of organic matter, and thus dehydrogenase activity can be an indicator of microbial redox system, which as a result can characterize soil microbial oxidation. Catalase (CAT) activities increased with increasing Cd concentrations in soil [30]. More critical Cd and Cu, with slight Zn pollution exhibited activation effect on soil enzyme such as urease, cellulase, alkaline phosphatase and polyphenol oxidase [31]. In this study, we found a decreasing trend in urease, alkaline phosphatase, dehydrogenase and catalase activity, as a result of increase in the levels of metal pollution. Less activation effect was shown in moderately contaminated soils but inhibitory effect was found in seriously polluted soil. At the same time when As content and urease, alkaline phosphatase, catalase activity displayed a negative correlation, there was a significant negative correlation in Cd and urease. That could restrain soil enzyme activity and even lead to inactivation. Thus As and other heavy metals inhibiting the enzyme may cause protein precipitation of enzyme inactivation, mainly because certain enzymes with thiol and amine molecules group were bonded to result in enzyme activity inhibition. Pagés D [32] found that inhibitory effect of heavy metal on enzyme may be due to their inhibition on microbial growth and reproduction which affects the synthesis and secretion of microbial enzymes in the body. The reasons for activating effect by heavy metals on enzyme production was considered as follows: (1) Cu, Zn and other metals increased activity as enzyme cofactor; (2) heavy metals stimulated microbial growth or reproduction and increased synthesis and secretion of enzymes in the body, thus boosting the enzyme activity; (3) some microorganisms surviving in soil polluted by high content of heavy metals become dominant microflora, which increases the synthesis and secretion of enzymes. In short, metabolism

of microbial communities and some of the enzyme showed activation effect and accelerated the transformation of the soil matrix correspondingly. However, because heavy metals can be absorbed by plants which it enriches, it is thus necessary for us to pay more attention to environmental evaluation of soil microbes and biological activity.

5. Conclusion

Study on heavy metals pollution and metabolism of soil microbes, activities of soil enzymes in iron tailing soil in the present research showed that Ni, As, Cu, Hg, Cd, V and Co pollution were discovered in the iron tailing area, with Ni and Cd pollution considered to be relatively more serious. Principal components analysis showed that the tailing pollution levels were closely related to metabolic properties of soil microbial communities. The significant change of metabolic characteristics between different soil microbial communities was observed along with increasing pollution levels. The change was mainly demonstrated by the differences in utilization of carbon by polymer and amino acids. In slightly and moderately polluted soils, the utilization of carbon sources by soil microbial communities was activated; while in heavily polluted soils, the carbon sources utilization was inhibited, furthermore in mildly and moderately polluted soils, the Shannon richness index (H), Simpson dominance index (1/D) and McIntosh index (U) were improved on the soil microbial community but they were inhibited in seriously polluted soils. The index of communities was most sensitive to heavy metal pollution, which could be used as an effective indicator of soil environmental quality in the similar iron tailing area. The activities of soil urease, alkaline phosphatase, catalase and dehydrogenase experienced declination with intensifying heavy metals pollution. The As content in soils impacted as negative correlation with urease, alkaline phosphatase, catalase activity.

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