

Soil Dehydrogenase Activity and Bacterial Community Diversity in the Water Level Fluctuation Zone of a Drinking Water Reservoir

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Soil microbial activity and community diversity in water level fluctuation zone (WLFZ) of the drinking water reservoir is still not well understood. In this context, soil dehydrogenase activity and bacterial community diversity of a drinking water reservoir was determined aiming to examine the water level fluctuation driving soil bacterial community activity and diversity across both vertical and longitudinal transects. BIOLOG method was employed to explore functional diversity of WLFZ soil bacterial community. The results shown that the highest dehydrogenase activity, 2.64 ig TF/g.24h, was found in the top of WLFZ, which was 2.67 times higher than that of the bottom. Meanwhile, the average well color development ($AWCD$)_{590nm}, species richness and Shannon's diversity of bacterial community associated with top of WLFZ was also significant higher than that of the bottom. The significant "site" and "WLFZ" revealed that the dehydrogenase activity, $AWCD$ _{590nm} and species richness varied among the sites within the WLFZ ($P < 0.01$). However, there were no significant two-way interactions for different sites ($P > 0.05$). Heatmap and principle component analyses (PCA) of bacterial community metabolic fingerprints suggested a significant discrimination soil bacterial community in the bottom, middle and top of three different sampling sites of WLFZ. The higher discriminate carbon substrates utilization by WLFZ soil bacterial community were cellobiose, hydroxy benzoic acid, tween 80, arginine, threonine, pyruvic acid methyl ester, ketobutyric acid, malic acid, glucosaminic acid and glycogen. These founding demonstrated that water level draw down affect soil microbial activity and bacterial community functional diversity in the water level fluctuation zone of the JIN PEN drinking water reservoir.

Key words: Bacterial community diversity, dehydrogenase activity, reservoir, water-level-fluctuating zone.

Water level fluctuation zone (WLFZ), an important area between the aquatic and the terrestrial ecosystem, is a critical region for soil biogeochemical processes of essential elements including carbon, nitrogen, phosphorous, and

sulfur and nutrients metabolic and transportation¹. During these processes, soil microbes largely drive geochemical cycling and their activities are crucial to the productivity of fluctuating belt environmental ecosystems with distinguishing hydrological regime². Soil bacterial community can be revealed by molecular methods such as real time-PCR, denatured gradient gel electrophoresis (DGGE), terminal restriction fragment length polymorphism (T-RFLP), microarray and pyrosequencing³⁻⁵. Meanwhile, BIOLOG method

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was widely used to determine soil microbial community functional diversity in restored forest^{3, 6}, agricultural⁷, wetland⁵ and river sediment⁴ ecosystem conditions. However, the report on bacterial community carbon metabolism in water level fluctuation zone (WLFZ) of reservoir is limited.

Drinking water reservoir is important for safety of urban water supply⁸. In the past few decades, WLFZ are increasingly attracting the researchers' attention because water level fluctuation can reduce the biodiversity with the changes of drying and re-flooding^{9,10}. Numerous reports have already revealed that the diversity of plant and animal communities was affected by water level fluctuation process. Previous studies have shown that water level fluctuation changes can alter the communities of plant^{9,10}, waterbird¹¹ species and ecological reconstruction^{12,13} of WLFZ. Ding *et al.*¹⁴ determined the heavy metal adsorption of purple soil in WLFZ of Three Gorges Reservoir, China. Recently, a field study conducted by Chen *et al.*¹³ demonstrated at flooding could decrease soil inorganic nitrogen and revegetation in the WLFZ of the Three Gorges Reservoir, and potentially improve water quality. At present, to our knowledge, the effects of water level fluctuation on the soil microbial activity and community in WLFZ of drinking water reservoir have been largely overlooked.

Consequently, the current study helps to close this gap. Here, this work was conducted to evaluate the soil microbial activity as dehydrogenase activity and bacterial community functional diversity in the water level fluctuation zone of the drinking water reservoir. The specific objectives were to (1) determine soil dehydrogenase activity and to (2) exam the soil bacterial community functional diversity in top, middle, bottom of three different sites (Site A, Site B and Site C) of WLFZ in the JIN PEN drinking water reservoir, the results will enrich our understanding of soil microbial functional characteristics in WLFZ of the drinking water reservoir.

MATERIALS AND METHODS

Study Site

The study drinking water reservoir, JIN PEN reservoir, is an important drinking water source

for Xi'an City. The reservoir is located in Zhouzhi County, Xi'an City, Shaanxi Province, northwest China (E108°112', N34°022'). The maximal depth is about 90-105 m, and the water volume is 2×10^8 m³. From September 2011 to June 2012, the water level fluctuated along the elevation from 558.99 m to 594.08 m. It is therefore the water level fluctuation zone is formed in the JIN PEN reservoir (Figure 1).

Sampling Description

In this work, field surveys were performed in July 6th 2012. The location (longitude and latitude) of sampling sites was determined by GPS device. The sampling sites were listed in Table 1. No plant or grass species harbored in the bottom of WLFZ, *Setaria viridis* and *Pinus tabulaeformis* were the dominant plant species grown in the middle and top of WLFZ in JIN PEN reservoir, respectively (Figure 1). In each sampling sites, three plots were selected. Each plot was 3×3 m. For each plot, four soil subsamples were randomly collected with a sterilized hand driven probes (5 cm diameter) to a depth of 15-20 cm. These four subsamples were fully mixed to act as a single soil sample per plot. The soil samples from bottom contained more sand than middle and top of the WLFZ. After being sieved through 2 mm mesh, the WLFZ soil samples were put into a small cooler with 8 °C and transported immediately to the laboratory of School of Environmental and Municipal Engineering, Xi'an University of Architecture & Technology (SEME-XUAT) within 12 h. Each soil sample was divided into two parts for soil dehydrogenase activity and bacterial community functional BIOLOG examination. The experiments were conducted in 48h after soil samples collection³.

Soil Dehydrogenase Activity Determination

To determine the soil microbial community activity, soil dehydrogenase activity was examined¹⁵. According to the method described by Guan¹⁶, Singh and Singh¹⁷ and little modification, 1 gram soil was added in the tube with 4 ml phosphate buffer and 1ml triphenylte trazolium chloride (TTC), and then the tubes were incubated for 24 h at 37 °C in the dark. Soil dehydrogenase enzyme can transfer TTC to 2, 3, 5-triphenyl formazan. WLFZ soil dehydrogenase activity was examined by reducing 2, 3, 5-triphenylte trazolium chloride. The absorption was spectrophotometer determined at 485 nm (UVmini-1204, SHIMADZU, Japan). The results were

expressed on the basis of the dry soil weight (oven-dried at 105° for 48 h) and the unit was expressed as µg TF/g.24h. The experiment was run in triplicate (n=3).

Soil Bacterial Community Functional Diversity Determination

To explore the WLFZ soil microbial community functional diversity, BIOLOG method was used to reveal bacterial community functional diversity. There are 31 different sole carbon sources located in the BIOLOG ECO micro plate (BIOLOG, Inc., Hayward, CA, USA) ¹⁸. According to the method by Garland ¹⁸ and Zhang *et al.* ³, 5.0 grams WLFZ soil (*d.w.*) was added into 45mL sterilized water, and then shook at 120 rpm for 30 min, diluted to 10⁻³ soil suspension in room temperature. 150 µl soil suspension was added into each well of ECO plate using eight pipetting device (Bio-Red, USA). All BIOLOG micro plates were then incubated at 28 °C in the dark for ten days ¹⁹. The BIOLOG data were recorded using the ELSVER reader every 12 hours interval at 590nm ^{3,18}. The soil bacterial community functional diversity indices were expressed as species richness and Shannon’s diversity²⁰. There are three replicates on each ECO plate ¹⁸. In this work, the data of 96 h incubation was used for *AWCD*_(590nm), community diversity index, heatmap and principle component analyses (PCA) ^{3,21}.

Statistical Analysis

All data were performed in triplicate (n =3) and analyzed by two-way analyses of variance (ANOVAs) using SAS statistic software (Version 8.1)(SAS Institute Inc., USA). Comparisons among

means were assessed by Tukey-Kramer HSD tests at a significance level (*P*<0.05). Heatmap (false color image) analyses of carbon sources utilization was run in the R software. Principle component analyses (PCA) was performed with SPSS (Version 16.0) for windows.

RESULTS AND DISCUSSION

Soil dehydrogenase activity

Soil dehydrogenase activity has been used as a good indicator of microbial activity ¹⁵. To date, little information is available on water level fluctuation zone (WLFZ) soil dehydrogenase activity of drinking water reservoir. In this study, soil dehydrogenase activity in the bottom of WLFZ is significant lower than that of the middle and top of WLFZ (*P*<0.05). As shown in Figure 2, the results shown that the highest dehydrogenase activity, 2.64 µg TF/g.24h, was found in the top of Site A WLFZ, 2.67 times higher than that of the lowest in the bottom Site C WLFZ. It is suggested that soil microbial community activity was decreased by longtime water logging. Soil dehydrogenase activity can be acted as a good determination of microbial oxidative activity. This result was consistent with Wang and Lu ²² reported that waterlogging decreased the soil microbial community oxidative activity, and this effect was enhanced with increasing waterlogging time. The most important reason might be the plant species diversity was suppressed by the waterlogging in the WLFZ area ^{9,23,24}. In this work, there is no plant grown in the bottom of WLFZ, because the

Table 1. Location (longitude and latitude) of the bottom, middle and top of water level fluctuation zone (WLFZ) in three different sampling sites (Site A, B, C) of the JIN PEN drinking water reservoir

Sampling sites	Water Level Fluctuation Zone (WLFZ)	Longitude (E)	Latitude (N)
Site A	Bottom (B)	108°11" 472 "	34°02" 302 "
	Middle (M)	108°11" 472 "	34°02" 292 "
	Top (T)	108°11" 452 "	34°02" 282 "
Site B	Bottom (B)	108°11" 342 "	34°02" 352 "
	Middle (M)	108°11" 322 "	34°02" 572 "
	Top (T)	108°11" 322 "	34°02" 342 "
Site C	Bottom (B)	108°11" 312 "	34°02" 132 "
	Middle (M)	108°11" 302 "	34°02" 212 "
	Top (T)	108°11" 312 "	34°02" 232 "

Table 2. Two-way ANOVA was performed to explore the effects of site and WLFZ on the soil dehydrogenase activity and bacterial functional community of the JIN PEN drinking water reservoir

Soil microbial community parameters	Site		WLFZ		Site ×WLFZ	
	<i>F</i> -value	<i>P</i> -ratio	<i>F</i> -value	<i>P</i> -ratio	<i>F</i> -value	<i>P</i> -ratio
Dehydrogenase activity	3.18	0.149 ^{NS}	21.88	0.007***	11.67	0.0001***
<i>AWCD</i> _(590nm)	1.95	0.256 ^{NS}	16.59	0.0116*	35.47	0.0001***
Richness diversity (<i>R</i>)	1.04	0.434 ^{NS}	62.98	0.0009***	10.11	0.0003***
Shannon's diversity (<i>H</i>)	2.97	0.162 ^{NS}	52.68	0.0013***	2.06	0.1346 ^{NS}

NS, not significant, $P > 0.05$, * $P < 0.05$, *** $P < 0.001$. The bold font value was not significant.

longtime waterlogging. Several studies have demonstrated the soil microbial community activity and enzyme activity were higher than that of bare land.

Soil bacterial community functional diversity

Soil bacterial community functional diversity can be used as bio-indicators for the soil quality of WLFZ in drinking water reservoir. Soil microbial community in the WLFZ in drinking water reservoir should be evaluated, because it's relationship with the drinking water quality. In the

Table 3. Higher discriminate carbon substrates in principle component analyses on the data of carbon substrates utilization by the bacterial community diversity harbored in water level fluctuation zone (WLFZ) of the JIN PEN drinking water reservoir

Carbon substrates located in BIOLOG ECO plate	PC1 score	PC2 score
D-Cellobiose	0.902	-0.003
D-Xylose	0.890	-0.027
N-Acetyl-D-glucosamine	0.742	0.090
2-Hydroxy benzoic acid	0.914	-0.038
Tween80	0.923	0.029
L-Arginine	0.917	0.060
L-Threonine	0.949	0.126
L-Phenylalanine	0.873	0.018
L-Asparagine	0.850	0.311
Pyruvic acid methyl ester	0.958	-0.049
Phenyl ethylamine	0.742	-0.323
Ketobutyric acid	0.915	0.028
D-Malic acid	0.947	0.079
Glucose-1-phosphate	0.740	0.274
Putrescine	0.730	-0.184
D-Glucosaminic acid	0.924	0.117
D-Galactonic acid lactone	0.788	-0.326
Glycogen	0.905	-0.230
Itaconic acid	0.885	-0.190

present study, bacterial community-level physiological profile (CLPPs) was examined by BIOLOG method. Heatmap revealed that different carbon sources were utilized by the soil bacterial communities in the bottom, middle and top of WLFZ in JIN PEN reservoir (Figure 3). Meanwhile, as shown in Figure 4, average well color development (*AWCD*), and diversity index were recorded. The species richness (*R*) and Shannon's diversity (*H*) were used to exhibit the functional diversity of WLFZ soil bacterial communities. The highest species diversity (*R*) was 27, which was in the top of WLFZ, the lowest was 2.67 in the bottom. The Shannon's diversity in top WLFZ of site C is 3.2, which is 2.29 times higher than that of bottom WLFZ of site A (Figure 4). The significant "WLFZ" and "Site ×WLFZ" indicates that the *AWCD* and

**Fig. 1.** The map of sampling sites in the water level fluctuation zone (WLFZ) in three different sampling sites (Site A, B, C) of the JIN PEN drinking water reservoir. Capital letters including B-A, B-B, B-C, M-A, M-B, M-C, T-A, T-B, T-C located in the figure represent Site A, Site B, Site C from the bottom, middle, top of the water level fluctuation zone (WLFZ), respectively

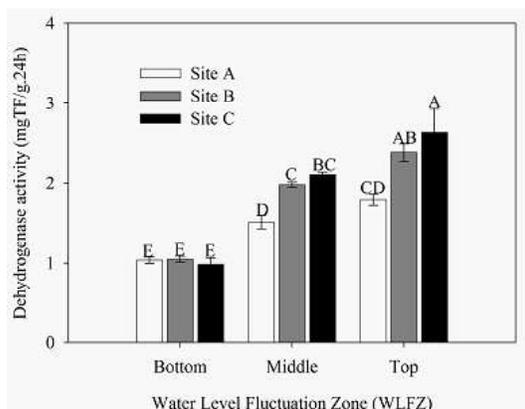


Fig. 2. Dehydrogenase activity of the soils from bottom, middle and top of the water level fluctuation zone (WLFZ) in three different sampling sites (Site A, B, C) of the JIN PEN drinking water reservoir. Data are the mean and standard error ($n=3$). Different capital letters above the bars indicate significant differences ($P<0.05$) assessed by Tukey-Kramer HSD

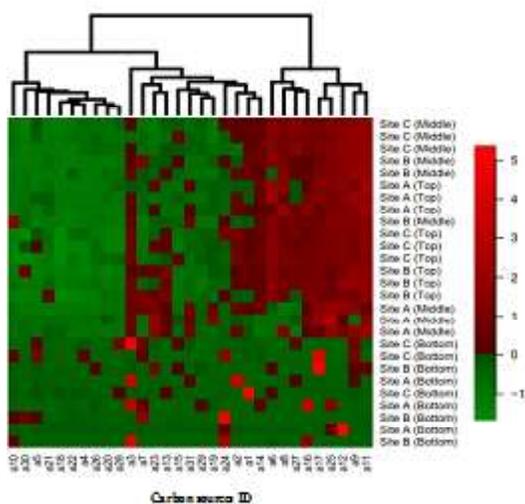


Fig. 3. Heatmap and cluster analysis of 31 sole carbon sources utilization by the soil bacterial communities from bottom, middle and top of the water level fluctuation zone (WLFZ) in three different sampling sites (Site A, B, C) of the JIN PEN drinking water reservoir. Carbon sources are in columns (carbon sources IDs are provided along the x axis) and rows are separate communities. Carbon sources are clustered based on similar utilization patterns across the WLFZ soil samples. The color code indicates soil bacterial community relative metabolic activity as average well color development, ranging from green (low activity) to red (high activity)

richness diversity varied among the WLFZ within the interaction. However, there was no significant two-way interaction in Shannon’s diversity (Table 2).

PCA of bacterial community BIOLOG ECO metabolic fingerprints revealed a significant discrimination the soil bacterial diversity in bottom, middle and top of three different sampling sites of WLFZ. The first and second principal component (PC1 and PC2), which explained 58.10% and 8.14% of the variance, respectively. Bottom was located in the third. PC1 mainly separates bottom and other two treatments (Figure 5). Carbon sources utilized by WLFZ soil bacterial community were determined. The higher discriminate carbon substrates in principle component analyses on the data of carbon source utilization was shown in table 3, including D-cellobiose, 2-hydroxy benzoic acid, tween80, L-arginine, L-threonine, pyruvic acid methyl ester, ketobutyric acid, D-malic acid, D-glucosaminic acid, and glycogen (Table 3).

BIOLOG method was widely used in detection soil microbial community functional diversity from several environmental conditions³⁻⁶. To our knowledge, there are few researches on the soil microbial metabolic activity in WLFZ of reservoir. The current study helps to close this gap. The current study shown that the soil microbial communities as dehydrogenase and community functional diversity were different in the bottom, middle and top of WLFZ in JIN PEN drinking water reservoir. The bottom of WLFZ was immersed in the water longer than middle and top, therefore, the enzyme metabolic and soil bacterial community diversity was suppressed, especially such as aerobic microorganism.

In the present study, it is also shown that the *AWCD*, and diversity index in the bottom of WLFZ was lowest. This result was similar with Yu *et al.*⁵ shown dramatic changes in microbial community function and structure along the successional gradient of coastal wetlands in Yellow River Estuary, it is might be that the water content of middle and top of WLFZ was better for aerobic soil bacterial activity. The heatmap and principle component analyses (PCA) also suggested that waterlogging in WLFZ can significantly shape soil bacterial community functional diversity. Although the soil bacterial community activity and functional diversity were determined in this work, the more

research is needed. Combined with high-throughput sequencing (454 pyrosequencing) and stable isotope labeling technique, the structure and functional of soil microbial species including ammonia oxidizing bacteria (AOB), ammonia oxidizing archaea (AOA) and methane-oxidizing bacteria (MOB) living in the WLFZ of drinking

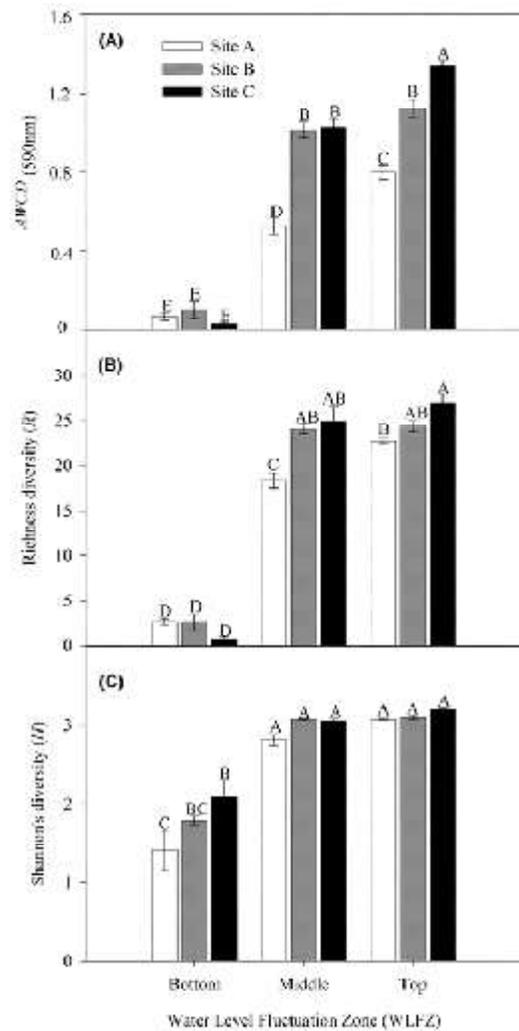


Fig. 4. (A) $AWCD_{(590nm)}$, (B) richness diversity (R) and (C) Shannon's diversity (H) of the diversity of soil bacterial community in the bottom, middle and top of the water level fluctuation zone (WLFZ) from three different sampling sites (Site A, B, C) of the JIN PEN drinking water reservoir. Data are the mean and standard error ($n=3$). Different capital letters above the bars indicate significant differences ($P<0.05$) assessed by Tukey-Kramer HSD

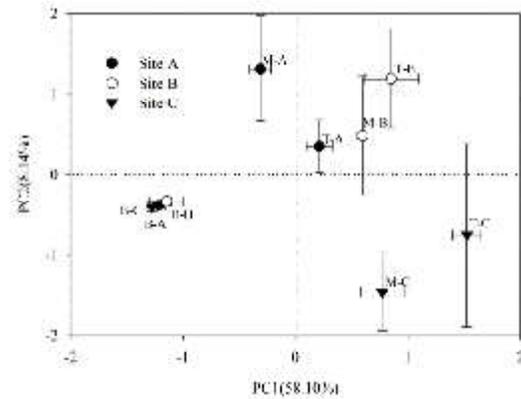


Fig. 5. Principal components analysis (PCA) of BIOLOG fingerprints of the bacterial community functional diversity in the soils from bottom, middle and top of the water level fluctuation zone (WLFZ) in three different sampling sites (Site A, B, C) of the JIN PEN drinking water reservoir. Data are the mean and standard error ($n=3$). PC1 and PC2 explain 58.10% and 8.14% of the total variance of the BIOLOG fingerprint data set, respectively. Capital letters including B-A, B-B, B-C, M-A, M-B, M-C, T-A, T-B, T-C located in the figure represent Site A, Site B, Site C from the bottom, middle, top of the water level fluctuation zone (WLFZ), respectively

water reservoir should be deeply investigated in the future.

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REFERENCES

1. Wang, H.D., Yang, Q., Pan, S.H., Ding, W.C., Gao, Y.L. Research on the impact of the water-

- level-fluctuation zone on landslide stability in the Three Gorges Reservoir area. *Appl. Mech. Mater.*, 2012; **188**: 37-44.
2. Paerl, H.W., Pinckney, J.L. A mini-review of microbial consortia: their roles in aquatic production and biogeochemical cycling. *Microb. Ecol.*, 1996; **31**: 225-247.
 3. Zhang, H.H., Tang, M., Chen, H., Zheng, C.L. Inoculation with ectomycorrhizal fungi affects microbial biomass and bacterial functional diversity in the rhizosphere of *Pinus tabulaeformis* seedlings. *Eur. J. Soil Biol.*, 2010; **46**: 55-61.
 4. Bushaw-Newton, K.L., Ewers, E.C., Ashley, D.J., Macavoy, S.E. Bacterial community profiles from sediments of the Anacostia River using metabolic and molecular analyses. *Environ. Sci. Pollut. Res.*, 2012; **19**: 1271-1279.
 5. Yu, Y., Wang, H., Liu, J., Wang, Q., Shen, T.L., Guo, W.H., Wang, R.Q. Shifts in microbial community function and structure along the successional gradient of coastal wetlands in Yellow River Estuary. *Eur. J. Soil Biol.*, 2012; **49**: 12-21.
 6. Wang, Y., Ouyang, Z.Y., Zheng, H., Wang, X.K., Chen, F.L., Zeng, J. Carbon metabolism of soil microbial communities of restored forests in Southern China. *J. Soil Sediment.*, 2011; **11**: 789-799.
 7. Nautiyal, C.S., Chauhan, P.S., Bhatia, C.R. Changes in soil physicochemical properties and microbial functional diversity due to 14 years of conversion of grassland to organic agriculture in semi-arid agro ecosystem. *Soil Till. Res.*, 2010; **109**: 55-60.
 8. Cong, H.B., Huang, T.L., Chai, B.B. A new mixing-oxygenating technology for water quality improvement of urban water source and its implication in a reservoir. *Renew Energ.*, 2009; **34**: 2054-2060.
 9. Bai, B.W., Wang, H.Y., Li, X.Y., Feng, Y.L., Zhi, L. A comparative study of the plant community of the future water-level-fluctuating zone and the natural water level fluctuating zone in the Three Gorges Reservoir. *J Southwest Agric Univ (Nat. Sci.)*, 2005; **27**: 684-691.
 10. Zhao, C.M., Chen, W.L., Huang, H.D., Tian, Z.Q., Chen, Y., Xie, Z.Q. Spatial pattern of plant species diversity in the inundation and resettlement region of the Three Gorges Reservoir. *Biodivers. Sci.*, 2007; **15**: 510-522.
 11. Francis, G., William, O., Samuel, K.O. Response of waterbird species to fluctuating water levels in tropical coastal wetlands. *Afr. J. Ecol.*, 2009; **48**: 637-643.
 12. Lu, Z.J., Li, L.F., Jiang, M.X., Huang, H.D., Bao, D.C. Can the soil seed bank contribute to revegetation of the drawdown zone in the Three Gorges Reservoir Region? *Plant Ecol.*, 2010; **209**: 153-165.
 13. Chen, Y., Cheng, X.L., Zhang, Y.L., Wang, Z.X., Zhang, Q.F. Soil nitrogen dynamics following short-term revegetation in the water level fluctuation zone of the Three Gorges Reservoir, China. *Ecol. Eng.*, 2012; **38**: 37-44.
 14. Ding, W.Q., Bao, B., Li, H., Song, Z.R. Heavy metal adsorption characteristics of purple soil in water-level-fluctuation zone of the Three Gorge Reservoir Region. *J. Ecol. Rural Environ.*, 2007; **23**: 40-42.
 15. Jorge-Mardomingo, I., Soler-Rovira, P., Casermeiro, M.A., María, T.C., Polo, A. Seasonal changes in microbial activity in a semiarid soil after application of a high dose of different organic amendments. *Geoderma*. 2013; **206**: 40-48
 16. Guan, S.Y. Soil enzyme and its research method. Beijing: Agriculture press, 1983.
 17. Singh, J., Singh, D. Dehydrogenase and phosphomonoesterase activities in groundnut (*Arachis hypogaea* L.) field after Diazinon, Imidacloprid and Lindane treatments. *Chemosphere.*, 2005; **60**: 35-42.
 18. Garland, J.L. Analytical approaches to the characterization of samples of microbial communities using patterns of potential C utilization. *Soil Biol. Biochem.*, 1996; **28**: 223-231.
 19. Classen, A.T., Boyle, S.I., Haskins, K.E., Overby, S.T., Hart, S.C. Community-level physiological profiles of bacteria and fungi: plate type and incubation temperature influences on contrasting soils. *FEMS Microbiol. Ecol.*, 2003; **44**: 319-328.
 20. Zak, J.C., Willig, M.R., Moorhead, D.L., Wildman, H.G. Functional diversity of microbial communities: a quantitative approach. *Soil Biol. Biochem.*, 1994; **26**: 1101-1108.
 21. Fierer, N., Lauber, C.L., Ramirez, K.S., Zaneveld, J., Bradford, M.A., Knight, R. Comparative metagenomic, phylogenetic and physiological analyses of soil microbial communities across nitrogen gradients. *ISME. J.*, 2012; **6**: 1007-1017
 22. Wang, X.C., Lu, Q. Effect of waterlogged and aerobic incubation on enzyme activities in paddy soil. *Pedosphere.*, 2006; **16**: 532-539.
 23. Guo, Q.S., Kang, Y., Hong, M., Jin, J.Q., Zhu, N.N., Nie, B.H., Wang, Z.Q. Responses of terrestrial plants in hydro-fluctuation belt of the Three Gorges Reservoir area to the first time flooding-drying habitat change. *Scientia Silvae*

- Sinicae.*, 2013; **49**: 1-9.
24. Schmieder, K., Dienst, M., Ostendorp, W., Jöhnk, K.D. Effects of water level variations on the dynamics of the reed belts of Lake Constance. *J Ecohydrology Hydrobiolog.*, 2004; **4**: 469- 480.