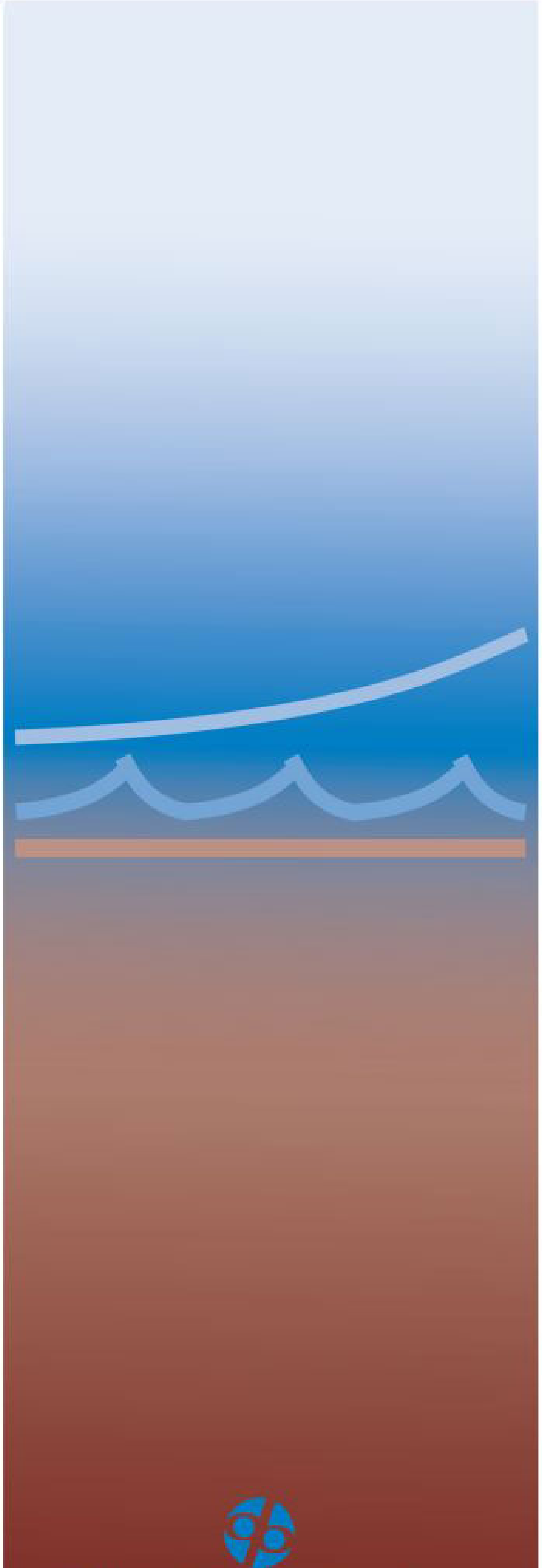


DES*tech* Publications, Inc.

# Journal of RESIDUALS SCIENCE & TECHNOLOGY

VOLUME 11, NUMBER 4, pp. 99-141 OCTOBER 2014
ISSN: 1544-8053

## Contents

Research

## Aim and Scope

The objective of the *Journal of Residuals Science & Technology* (JRS&T) is to provide a forum for technical research on the management and disposal of residuals from pollution control activities. The Journal publishes papers that examine the characteristics, effects, and management principles of various residuals from such sources as wastewater treatment, water treatment, air pollution control, hazardous waste treatment, solid waste, industrial waste treatment, and other pollution control activities. Papers on health and the environmental effects of residuals production, management, and disposal are also welcome.

## Editor-in-Chief

P. Brent Duncan
*Department of Biology*
*University of North Texas*
*Denton, TX, USA*
pduncan@unt.edu

## Editorial Advisory Board

Muhammad Abu-Orf
*AECOM, USA*
mohammad.abu-orf@aecom.com

Steve Dentel
*University of Delaware, USA*
dentel@udel.edu

Richard Dick
*Cornell University, USA*
rid1@cornell.edu

Guor-Cheng Fang, Ph.D.
*Hungkuang University, Taiwan*
gcfang@sunrise.hk.edu.tw

Robert Hale
*Virginia Iustitute of Marine Science, USA*
hale@vims.edu

Paul F. Hudak
*University of North Texas, USA*
hudak@unt.edu

Blanca Jimenez Cisneros
*Inst. de Ingenieria, UNAM, Mexico*
bjc@mumas.iingen.unam.mx

Julia Kopp
*Technische Universitat*
*Braunschweig, Germany*
j.kopp@tu-bs.de

Uta Krogmann
*RutgersUniversity, USA*
krogmann@aesop.rutgers.edu

D. J. Lee
*National Taiwan University, Taiwan*
djlee@ntu.edu.tw

Giuseppe Mininni
*Via Reno 1, Italy*
mininni@irsa.rm.cnr.it

John Novak
*Virginia Tech, USA*
jtnov@vt.edu

Nagaharu Okuno
*The University of Shiga Prefecture,*
*Japan*
okuno@ses.usp.ac.jp

Jan Oleszkiewicz
*University of Manitoba, Canada*
oleszkie@ms.umanitoba.ca

Banu Örmeci
*Carleton University, Canada*
banu_ormeci@carleton.ca

Ian L. Pepper
*University of Arizona, USA*
ipepper@ag.arizona.edu

Ioana G. Petrisor
*Co-Editor-in-Chief*
*Environmental Forensics Journal, USA*
Environmental.Forensics@gmail.com

Bob Reimers
*Tulane University, USA*
rreimers@tulane.edu

Dilek Sanin
*Middle East Technical University,*
*Turkey*
dsanin@metu.edu.tr

Mike Switzenbaum
*Professor Emeritus*
*Marquette University, USA*
michael.switzenbaum@marquette.edu

Heidi Snyman
*Golder Associates Africa (Pty) Ltd.,*
*South Africa*
hsnyman@golder.co.za

Ludovico Spinosa
*Consultant at Commissariat*
*for Env. Energ. in Region,*
*Puglia, Italy*
ludovico.spinosa@fastwebnet.it

P. Aarne Vesilind
*Bucknell University, USA*
aarne.vesilind@gmail.com

Doug Williams
*California Polytechnic State*
*University, USA*
wmsengr@thegrid.net

**JOURNAL OF RESIDUALS SCIENCE & TECHNOLOGY**—Published quarterly—January, April, July and October by DEStech Publications, Inc., 439 North Duke Street, Lancaster, PA 17602.

Indexed by Chemical Abstracts Service. Indexed/abstracted in Science Citation Index Expanded. Abstracted in Current Contents/Engineering, Computing & Technology. Listed in ISI Master Journal.

Subscriptions: Annual $219 per year. Single copy price $60. Foreign subscriptions add $45 per year for postage.

(ISSN 1544-8053)

DES*tech* Publications, Inc.
439 North Duke Street, Lancaster, PA 17602-4967, U.S.A.

*©Copyright by DEStech Publications, Inc. 2014—All Rights Reserved*

# C O N T E N T S

## Research

# Bioremediation of PAHs from Contaminated Soils by *Festuca aroundiacea* in the Presence of *Bacillus licheniformis* and *Bacillus mojavensis*

SOMAYEH ESKANDARY[1], MEHRAN HOODAJI[2,*], AREZOO TAHMOURESPOUR[3] and ATOUSA ABDOLLAHI[4]
[1]*PhD student of Soil Science, Department of Soil Science, Islamic Azad University, Isfahan (Khorasgan) Branch, Isfahan, Iran*
[2]*Associate Professor of Soil Science, Department of Soil Science, Islamic Azad University, Isfahan (Khorasgan) Branch, Isfahan, Iran*
[3]*Assistant Professor of Microbiology, Department of Basic Medical Science, Islamic Azad University, Isfahan (Khorasgan) Branch, Isfahan, Iran*
[4]*Assistant Professor of Chemistry, Department of Environmental Chemistry, Islamic Azad University, Isfahan (Khorasgan) Branch, Isfahan, Iran*

**ABSTRACT:** Bioremediation of PAHs from oil-contaminated soils was investigated. *Bacillus licheniformis* and *Bacillus mojavensis*, were inoculated in rhizospheric soil with *Festuca aroundiacea* in a greenhouse study. The concentration of PAHs in the rhizosphere of soil inoculated with both bacteria were significantly ($p < 0.05$) reduced compared to the non-inoculated soil. PAH concentrations in shoots were largely independent of those in soil, roots, or rhizosphere soil. In the soil inoculated with both bacteria, the concentration of PAHs in roots and shoots of the plant was less than for other treatments. Results showed that the plant-promoted biodegradation was the predominant contribution to remediation of soil PAHs.

## INTRODUCTION

PAHs are aromatic compounds with two or more benzene rings. Usually, they are formed during the thermal degradation of organic molecules and similar compounds [1]. The common sources of these compounds in the environment can be human activities, forest and grassland fires, oil spills, volcanoes and industries [2]. The PAHs compounds are known as toxic, mutagenic and carcinogenic pollutants [3]. They are not easily eliminated from the environment under normal circumstances. If the molecular weight increases, their resistance to degradation will increase. Due to their widespread presence in the air, soil and sediments, they have attracted attention. These compounds may be eliminated from the environment in one of the following ways: evaporation, light oxidation, chemical oxidation, superficial adsorption of soil particles and leaching [4].

These methods are usually expensive, ineffective, costly and time consuming and transmission of contamination from one phase to another. Among all the studied methods used for the elimination of such compounds from the environment, bioremediation and phytoremediation are processes that requires the least amount of energy and chemical substance. They are degrading the dangerous pollutants to less dangerous ones [5].

Maiti *et al.* (2013) isolated and identified one strain of *Bacillus* from oil contaminated soil in India and found that it could mineralize antheracene, fluoranthene, pyrene and benzo (a) pyrene [6].

Toledo (2006) isolated fifteen bacterial strains from solid waste oil samples for their capacity to grow in the presence of naphthalene, phenanthrene, fluoranthene, or pyrene as the sole carbon sources. The isolates were identified by 16S rDNA sequence, and results showed that the strains belonged to the genera *Bacillus*, *Bacillus pumilus* (eight strains), and *Bacillus subtilis* (two strains) [7].

In fact, phytoremediation is a technology that acts based on the natural activities occurring in the soil. These activities include symbiotic relationships between plants, microorganisms and the environment. Phytoremediation has advantages such as plant's com-

---

*Author to whom correspondence should be addressed.
E-mail: M_Hoodaji@khuisf.ac.ir; Tel: +98 9131172651

1544-8053/14/04 099-07

plex root system that occupies a large amount of the soil and supports a large population of bacteria in the rhizosphere. Its secretions can directly affect the activity of the bacterial population in the rhizosphere and eliminate pollution. In fact, one of the best methods for cleansing the oil-contaminated soils is phytoremediation. For this purpose, plants should be used which are more compatible and flexible in harsh conditions caused by hydrocarbons [8].

Research has shown that plant roots have a great effect on PAH decomposition in such a way that a high level of decomposition happens in the vicinity of the plant roots [9].

Although abiotic factors such as pH, moisture, oxygen and available nutrients can affect the decomposition of PAHs, microbial processes notably have a greater share in the decomposition of these compounds [10]. Petroleum hydrocarbons absorb specific populations of decomposing bacteria and cause the expansion of decomposing bacterial populations [1].

Banks *et al.* (2003) stated that a large range of grasses and legume herbs with their symbiotic bacteria have shown increased removal of oil pollutants from the soil [8].

Some studies have also shown that plant affect uptake of lipophilic contaminants from soils, and results show that the plant lipids are the major factor for the differences in plant lipophilic compounds uptake such as dieldrin, heptachlor, aldrin and heptachlorepoxide [11].

However, there is still a lake of direct evidence to evaluate the plant uptake, accumulation and subsequent translocation of PAHs. Although, several studies have revealed the uptake and accumulation of PAHs from the atmosphere as a result of the deposition PAHs particle bound compounds on the leaf cuticle [11], uptake and accumulation of PAHs by plant in oil-contaminated soils are still under study, and information is scant on correlations between plant PAH concentrations and PAHs translocation in plants.

In this study, *Festuca arundinacea* was inoculated with two indigenous bacteria isolated from oil-contaminated soil. The species of bacteria for inoculation were *Bacillus licheniformis ATHE9* and *Bacillus mojavensis ATHE13* and their effect on the removal of hydrocarbons from the plant's rhizosphere was studied.

## MATERIALS AND METHODS

### Sampling

Soil samples were collected from contaminated soils around the tanks of the Isfahan Oil and Gas Refinery that occasionally collected and were piled up to reduce the risk of ignition and occurrence of fire with pH 7 and 4.7% organic matter. The 10 following polycyclic aromatic hydrocarbons were studied: naphthalene (NAP), acenaphthene (ANA), acenaphthylene (ANY), phenanthrene (PHE), anthracene (ANT), benzo[a]anthracene (BaA), chrysene (CHR), benzo[a]pyrene (BaP), dibenzo[a,h]anthracene (DBahA), benzo[ghi]perylene (BPY). Before transporting to the greenhouse, to ensure the homogeneity of the treatments, soils were sieved through 3 mm mesh and mixed.

### Isolation and Identification of PAHs Degrading Bacteria

The enrichment, isolation, morphological and physiological characteristics, analysis of 16S rDNA sequence and evaluation PAHs-degradation potential of degrading bacteria in oil-contaminated soil samples were conducted using Zhang *et al.* (2009) methods [12]. Then two bacterial isolates with more optical density and degradation ability of PAHs were selected for further studies.

### Seed Inoculation with Bacteria

From two selected bacteria (*Bacillus licheniformis* and *Bacillus mojavensis*) an inoculum liquid with the concentration of McFarland standard No. 3 was prepared and seeds were placed in inoculum liquid plates for 5 to 6 hours before they cultured. McFarland standards were used as a reference to adjust turbidity of bacterial suspensions so the number of bacteria will be within a given range for standardized microbial testing. After that in the seedling stages, one mL of the same inoculum liquid were inoculated to the plant after 3 weeks [13].

### Experimental Procedure

An equal amount of contaminated soil was added to each pot (7 kg) and twenty seeds of *Festuca aroundiacea* inoculated with bacteria were planted in the pots. This study was designed as a completely randomized block factorial with three replications. The plants were grown under controlled conditions for 3 months. They were exposed to light for 18 hours with a temperature range of 2–30°C. The relative humidity was fixed at 70%. After 3 months the rhizosphere soil around the roots was gently collected and transferred to the laboratory for PAHs extraction. The plant roots and shoots were carefully rinsed with distilled water, blotted dry with a tissue paper and frozen at –20ºC prior to determination of the PAHs concentration [14].

### PAHs Analyses of Soil and Plant Samples

In order to investigate the PAH concentration, 5 g of soil and plant samples were passed through a 2 mm sieve and 5 g of active sodium sulfate was added. They were then extracted by a mixture of 150 ml of acetone and dichloromethane with a volume ratio of 1:1 using a soxhlet apparatus. After its volume was reduced with rotary evaporation and then dried with nitrogen gas, they were kept in the freezer at –20°C until injection time [14].

At the time of injection of the soil samples to GC-FID (Agilent 7890 A), certain concentrations of m-Terphenyl as internal standard were added to all samples (Rodrigo *et al.* 2009). To measure PAHs with a GC-FID device, a HP-5 column (30 m × 0.25 mm I.D. 0.25 µm film thickness, Agilent) was used. GC-FID device was programmed with inlet temperature of 260°C and $N_2$ carrier gas flow of 1.5 ml $min^{-1}$.

The initial temperature of 70°C reached 290°C with the gradient of 5°C. Subsequently, it reached 305°C with the gradient of 1°C $min^{-1}$. Detector temperature was 270°C in this program.

Analysis of polycyclic aromatic hydrocarbons in plant samples were carried out by an Agilent GC Model 6890 coupled to a quadrupole mass spectrometer (5975 C). The Phenanteren $d_{10}$ was added in specific concentration as an internal standard to all samples. The system was operated in electron impact mode (EI, 70 eV). A HP-5 MS column (30 m × 0.25 mm I.D. 0.25 µm film thickness, Agilent) and a temperature programming were used in order to get the proper GC separation. The temperature started from 80ºC with a 5 min hold time, it was then increased to 150ºC at a rate of 10ºC $min^{-1}$. It was then increased to 300ºC at a rate of 5ºC $min^{-1}$, keeping the final temperature for 5 min. Injection was performed in the split less mode. Helium gas was used as a carrier gas at a constant flow rate of 1.5 ml $min^{-1}$.

The injector and transfer line temperatures were 250 and 280ºC, respectively. Ions were selected after considering the total ion chromatogram of solution of compounds. The ions were divided into four groups. Peak detection and integration were carried out using Chemstation software and AMDIS (automated mass spectral deconvolution and identification system).

### Statistical Analysis

Statistical analyses were performed using SAS software and the comparison of means was done with Duncan test at 5% level. The *Festuca* inoculated with *Bacillus licheniformis ATHE9* (F1), *Festuca* inoculated with *Bacillus mojavensis ATHE13* (F8), *Festuca* inoculated with both bacteria (F1,8) and *Festuca* without inoculation (F0) as control treatment.

## RESULTS AND DISCUSSION

The isolated bacterial strains of *ATHE9* and *ATHE13* were identified as *Bacillus licheniformis* and *Bacillus mojavensis* according to its 16S rDNA sequence as well as biochemical characteristics. They are gram-positive short rods, motile and spore forming, aerobic, oxidase positive, catalase positive, manitol positive, citrate positive.

*Bacillus licheniformis strain ATHE9* and *Bacillus mojavensis strain ATHE13* could utilize naphthalene, phenanthrene, anthracene, pyrene and other PAHs as sole carbon sources. The degradation efficiencies were examined by GC-Mass and the results showed that these isolates could remove PAHs (the initial concentration of 12.8 mg $L^{-1}$) in 6 days at 30°C and pH 7.3, accordingly these two isolates were selected to inoculate the plant. The Accession No. of *Bacillus licheniformis ATHE9* and *Bacillus mojavensis ATHE13* in gene bank are KC329470.1 and KC469987.1 respectively.

### Concentrations of PAHs in Soil

Before transferring the soil in to the pots and applying treatments, the hydrocarbon concentrations of the soil were measured. The chemical and physical properties of soil and concentrations of PAHs were showed in Table 1.

The concentrations of these hydrocarbons in the soil are much higher than the standard range of 50 to 1100 µg $kg^{-1}$ [15]. Therefore, these results confirm the needs for cleanup the soil from hydrocarbons. Furthermore, leaving this soil in non-isolated conditions causes contamination of groundwater and air pollution because these compounds spread in water and air based on their solubility coefficient and steam pressure.

After 3 months, the concentrations of 10 PAHs in soil decreased significantly ($p < 0.05$). More than 77, 82, 68, 45, 11, 21, 0.6, 32, 4 and 12 percent of naphthalene, acenaphthene, acenaphthylene, phenanthrene, anthracene, benzo[a]anthracene, chry-

***Table 1. Physical and Chemical Properties and Concentration of Polycyclic Aromatic Hydrocarbons in Contaminated Soil.***

| Characteristic | Amount |
|---|---|
| pH | 7.24 ± 0.04 |
| EC (dS.m$^{-1}$) | 3.4 ± 0.25 |
| CEC (meq.100 g$^{-1}$) | 7 ± 1 |
| OM (%) | 4.6 |
| Clay (%) | 16.6 |
| Sand (%) | 75 |
| Silt (%) | 8.4 |
| FC water content (%) | 22.4 |
| Naphthalene (mg.kg$^{-1}$) | 16 ± 6 |
| Acenaphthene (mg.kg$^{-1}$) | 18 ± 4 |
| Benzo[a]anthracene (mg.kg$^{-1}$) | 22 ± 8 |
| Benzo[a]pyrene (mg.kg$^{-1}$) | 5 ± 1/3 |
| Benzo[ghi]perylene (mg.kg$^{-1}$) | 2/5 ± 1 |
| Acenaphthylene (mg.kg$^{-1}$) | 32 ± 9 |
| Antrhracene (mg.kg$^{-1}$) | 4/5 ± 2 |
| Phenanthrene (mg.kg$^{-1}$) | 6 ± 2 |
| Chrysene (mg.kg$^{-1}$) | 16 ± 6 |
| Dibenzo[a,h]anthracene (mg.kg$^{-1}$) | 8 ± 2 |

sene, benzo[a]pyrene, dibenzo[a,h]anthracene and benzo[ghi]perylene, respectively, were removed from the non- inoculated soil (F0 in Figure 1) as the treatment with lowest reduction during the experimental period. The effectiveness of each treatment in reducing the hydrocarbon concentrations in rhizosphere are shown in Figures 1 and 2.

As seen in Figures 1 and 2, the concentration of hydrocarbons in the rhizosphere of the treated samples greatly decreased in comparison with the initial contaminated soil (Table 1). In the rhizosphere, hydrocarbons with 2 or 3 benzene rings were more degraded compared to heavier hydrocarbons with more rings. Heavy hydrocarbons were less exposed to microorganisms degradation because of their lower solubility in aqueous phase and higher adsorption on soil surfaces [16].

Figure 1 shows that the decomposition of 2 and 3-ring hydrocarbons in *Festuca* and *Bacillus mojavensis ATHE13* (F8) treatment was much higher than other samples.

Given these facts, it can be stated that the *Bacillus mojavensis ATHE13* has high ability for improving the conditions of plant roots and it causes higher decomposition of hydrocarbons with less rings and eliminates them from the rhizosphere. Furthermore, the difference in behavior of the bacteria in degenerating various hydrocarbons (in terms of rings number) is because of the unique differences of each bacterium. Each bacterium based on its specific enzyme systems acts specifically in the decomposition of some types of hydrocarbons [17].

Figure 2 show that sample inoculated with both bacteria has lower concentration of hydrocarbons with 4, 5 and 6 rings. These results suggest that the presence of both bacteria together is effective in the decomposition of heavier hydrocarbons with higher number of

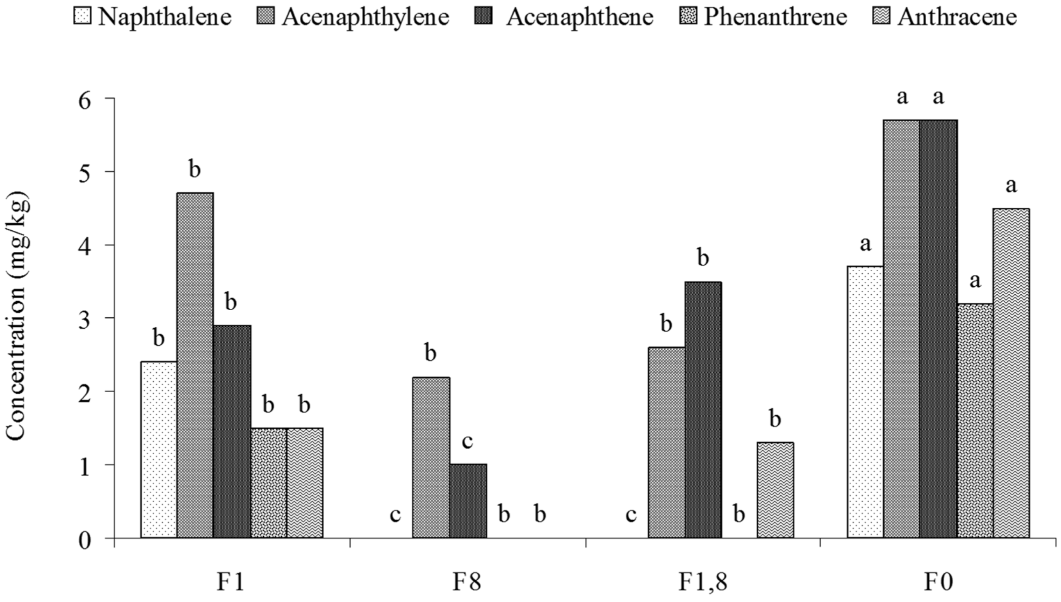


***Figure 1.** The concentration of polycyclic (2 and 3 rings) aromatic hydrocarbons in the treated rhizosphere. Festuca inoculated with Bacillus licheniformis ATHE9 (F1), Festuca inoculated with Bacillus mojavensis ATHE13 (F8), Festuca inoculated with both bacteria (F1,8) and Festuca without inoculation (F0).*

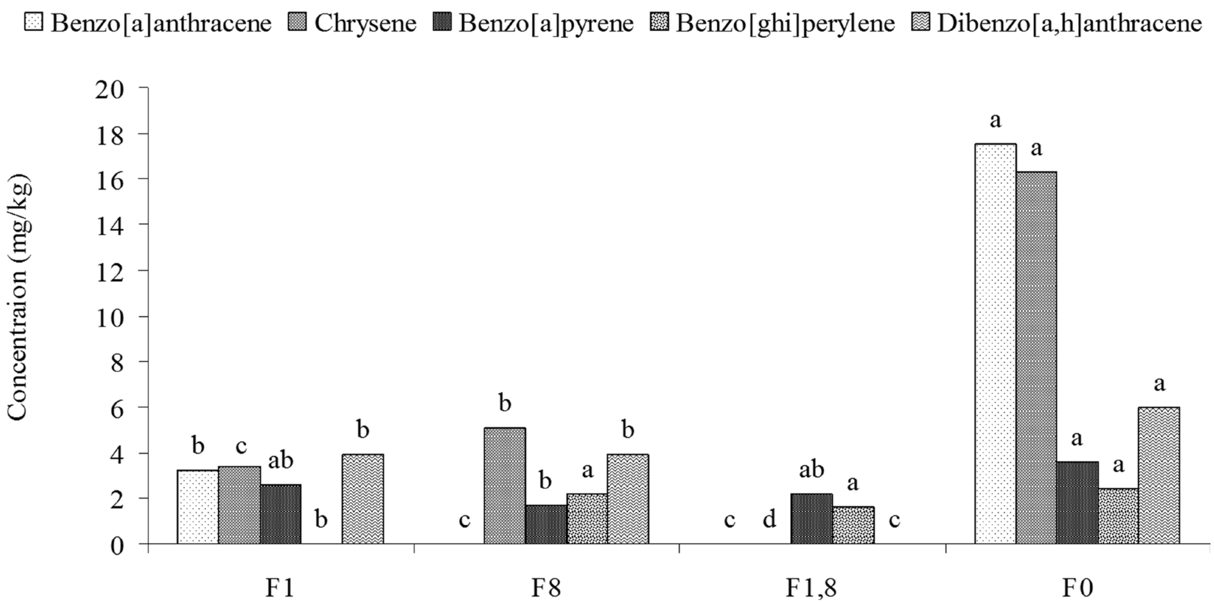


***Figure 2.** Comparison of the concentration of polycyclic (4, 5 and 6 rings) aromatic hydrocarbons in the treated rhizosphere. Festuca inoculated with Bacillus licheniformis ATHE9 (F1), Festuca inoculated with Bacillus mojavensis ATHE13 (F8), Festuca inoculated with both bacteria (F1,8) and Festuca without inoculation (F0).*

benzene rings. Haritash and Kaushik in 2009 reported similar results [10].

Additionally, the results also showed that the concentrations of hydrocarbons in the treatment of *Festuca* without inoculation and inoculation with *Bacillus licheniformis ATHE9* were significantly higher than other treatments ($p < 0.05$) so *Bacillus mojavensis ATHE13(F8)* and also both bacteria (F1,8) will make the pollution condition more tolerable for the plants and the rhizosphere of the plant is able to degrade more PAHs. Su and Zhu in 2007 also presented such results [18]. According to the results obtained in this study, it can be stated that the mechanism of phytoremediation, which involves the plant and its dependent microbial activities in the rhizosphere, is greatly capable of decomposing and eliminating PAHs from soil environments.

## Concentrations of PAHs in Plants

Concentrations of Dibenzo[a,h]anthracene and Benzo[ghi]perylene in roots and shoots were not detectable. But concentrations of other PAHs in roots were higher than shoots (Table 2). Acenaphthylene, Acenaphthene, Phenanthrene, Anthracene and Benzo[a]anthracene concentrations in roots increased proportionally with those in rhizosphere soils. With increasing log $K_{ow}$ values of the compounds the PAH concentrations in roots decreased. Similar results for other contaminants in plant roots were reported. Root

***Table 2. Root and Shoot PAH Concentrations (mg kg$^{-1}$, dry weight).***

| | | NAP | ANY | ANA | PHE | ANT | BaA | CHR | BaP |
|---|---|---|---|---|---|---|---|---|---|
| Roots | F1 | 0.2 ± 0.02 | 0.1 ± 0.04 | 0.06 ± 0.01 | 0.10 ± 0.01 | 0.09 ± 0.004 | 0.03 ± 0.004 | 0.04 ± 0.003 | 0.03 ± 0.002 |
| | F8 | 0.16 ± 0.03 | 0.11 ± 0.04 | 0.06 ± 0.008 | 0.11 ± 0.09 | 0.09 ± 0.002 | 0.03 ± 0.003 | 0.03 ± 0.008 | 0.02 ± 0.009 |
| | F1,8 | 0.14 ± 0.07 | 0.08 ± 0.007 | 0.04 ± 0.01 | 0.08 ± 0.02 | 0.06 ± 0.005 | 0.02 ± 0.007 | 0 | 0.05 ± 0.002 |
| | F0 | 0.20 ± 0.05 | 0.12 ± 0.08 | 0.09 ± 0.008 | 0.12 ± 0.08 | 0.12 ± 0.02 | 0.18 ± 0.08 | 0.06 ± 0.009 | 0.09 ± 0.006 |
| Analysis of variance | | NS | * | * | ** | ** | ** | ** | ** |
| Shoots | F1 | 0.19 ± 0.02 | 0.11 ± 0.03 | 0.06 ± 0.005 | 0.11 ± 0.05 | 0.09 ± 0.03 | 0.10 ± 0.008 | 0.03 ± 0.005 | 0.03 ± 0.007 |
| | F8 | 0.19 ± 0.01 | 0.08 ± 0.01 | 0.04 ± 0.005 | 0.09 ± 0.06 | 0.06 ± 0.006 | 0.07 ± 0.009 | 0 | 0 |
| | F1,8 | 0.1 ± 0.02 | 0.07 ± 0.004 | 0.04 ± 0.01 | 0.07 ± 0.04 | 0.08 ± 0.007 | 0.02 ± 0.006 | 0.02 ± 0.009 | 0.02 ± 0.003 |
| | F0 | 0.20 ± 0.07 | 0.11 ± 0.03 | 0.15 ± 0.08 | 0.10 ± 0.03 | 0.05 ± 0.002 | 0.15 ± 0.005 | 0.10 ± 0.001 | 0.06 ± 0.005 |
| Analysis of variance | | NS | NS | ** | * | * | ** | * | * |

**$P < 0.01$; *$P < 0.05$; NS, not significant.

accumulations of these PAHs increased with their concentrations in soil. By contrast, the lipids in roots were usually the major reservoir for highly water insoluble contaminants [19].

Results indicated that inoculation treatments had more effect on removal of PAHs. In the treatment inoculated with both bacteria, accumulation of PAHs in the shoots and roots was less than others. This shows that bacteria have reduced the hydrocarbon concentrations in the rhizosphere that is why plants uptake them less (Figure 1 and 2).

However, the concentrations of the ten PAHs in Festuca shoots showed significant differences with various treatments and were correlated with those in the roots. Wild *et al.*, (2005) indicated that phenanthrene and anthracene translocate slowly into plant roots, only up to 1500 μm in length over a 56-day period [20]. Although there is a lack of information on root uptake of PAHs, this study showed accumulation of PAHs in roots. Several studies suggested that the root uptake of lipophilic organic compounds can be in correlation with root lipid contents [21].

Shoot uptake of PAHs also was enhanced along with the increase of their concentrations in root. The concentrations of PAHs in shoots were statistically less than in roots. Howsam *et al.*, (2001) suggested that uptake of lipophilic organic compounds from the atmosphere by shoots is dependent on the plant lipid contents and plant surface area [22].

## CONCLUSION

Phytoremediation is one of the best ways to eliminate PAHs from the soil. In the this study, *Festuca* was inoculated with two types of *Bacillus*. The results of the study indicate that phytoremediation can be effective in the removal of PAH contamination from soil. Moreover, the results showed that inoculation of bacteria to plants could play an important role in the rate of elimination of hydrocarbons. There were different rates of PAHs uptake to roots or shoots from soil. PAHs may be taken up by roots via passive processes to subsequently accumulate into the root organic matter. The accumulation of PAHs was well correlated with PAH concentrations in rhizosphere soils. Because of the low PAH mass transport rates in soil, it was difficult for PAHs to translocate from rhizosphere soils to roots. Therefore, for soils contaminated with PAHs, both the plant uptake capacity and the PAH mass transport rate must be taken into account to assess the phytoremediation efficiency. Results from the present study indicated that contributions of plant uptake and rhizosphere effect to remove PAHs from soils are significant and rhizosphere effect and plant uptake have important role to remove PAHs from contaminated soils.

## REFERENCES

1. Riser-Roberts, E., “Remediation of petroleum contaminated soils: biological, physical and chemical processes”, Boca Raton, FL. Lewis Publishers. 1998. CRC press LLC.
2. Kostecki, P.T. and Calabrese E.J., “Hydrocarbon Contaminated Soils”, Lewis Publishers, Chelsea, MI, 1993, Vol. 3.
3. Han, M.J., Choi, H.T., Song, H.G., “Degradation of phenanthrene by *Trametes versicolor* and its laccase, *J. Microbiol.,* Vol. 42, No. 2, 2004, pp. 94–8.
4. Newman, L.A., Strand, S.E., Choe, N., Duffy, J., Ekuan, G., Ruszaj, M., Shurtleff, B.B., Wilmoth, J., Heilman, P., Gordon, M.P., “Uptake and biotransformation of trichloroethylene by hybrid poplars”, *Environ. Sci. Technol.*, Vol. 31, No. 4, 1997, pp. 1062–1067.
5. Siciliano, S.D. and Germida, J.J., “Mechanisms of phytoremediation: Biochemical and ecological interactions between plants and bacteria”, *Environ. Rev,* Vol. 6, 1998, pp. 65–79.
6. Maiti, A., Das, S., Bhattacharyya, N., “High gelatinase activity of a newly isolated polycyclic aromatic hydrocarbon degrading bacteria *Bacillus weihenstephanensis* strain AN1”, *Journal of Pharmacy Research,* Vol. 6, No. 1, 2013, pp. 199–204.
7. Toledo, F.L., Calvo, C., Rodelas, B., Gonzalez-Lopez, J., “Selection and identification of bacteria isolated from waste crude oil with polycyclic aromatic hydrocarbons removal capacities”, *Syst. Appl. Microbiol.,* Vol. 29, 2006, pp. 244–252.
8. Banks, M.K., Schwab, P., Liu, B. Kulakow, P.A., Smith, J.S., Kim, R., “The Effect of Plants on the Degradation and Toxicity of Petroleum Contaminants in Soil: A Field Assessment”, *Adv. Biochem. Engin./Biotechnol.,* Vol. 78, 2003, pp. 75–96.
9. Doyle, E., Muckian, L., Hickey, A.M., Clipson, N., “Microbial PAH degradation”, *Adv. Appl. Microbiol.*, Vol. 65, 2008, pp. 27–66.
10. Haritash, A.K. and Kaushik, C.P., “Biodegradation aspects of poly aromatic hydrocarbons (PAHs)”, *J. Hazard. Mater.,* Vol. 169, No. 1–3, 2009, pp. 1–15.
11. Gao, Y.Z. and Zhu, L.Z., “Plant uptake, accumulation and translocation of phenanthrene and pyrene in soils”, *Chemosphere,* Vol. 55, 2004, pp. 1169–1178.
12. Zhang, G.Y., Ling, J.Y., Sun, H.B., Luo, J., Fan, Y.Y., Cui, Z.J., “Isolation and characterization of a newly isolated polycyclic aromatic hydrocarbons-degrading *Janibacter anophelis* strain JY11”, *J. Hazard. Mater.,* Vol. 172, No. 2-3, 2009, pp. 580–6.
13. Hamdi, H., Benzarti, S., Manusadzianas, L., Aoyama, L., Jedidi, N., “Bioaugmentation and biostimulation effects on PAH dissipation and soil ecotoxicity under controlled conditions”, *Soil Biol. Biochem.,* Vol. 39, 2007, pp. 1926–1935.
14. Su Y.H. and Zhu Y.G., “Uptake of selected PAHs from contaminated soils by rice seedling (Oryza sativa) and influence of rhizosphere on PAH distribution”, *Environ. Pollut.,* Vol. 155, No. 2, 2008, pp. 359–65.
15. Peijnenburg, W.J.G.M., “Fate of Contaminants in Soil”, In: P. Doelman, H. Eijsackers (Eds). *Vital soil. Function, value and properties.* Elsevier, Amsterdam, the Netherlands, 2004, pp. 245–280.
16. Soleimani, M., Afyuni, M., Hajabbasi, M., Nourbakhsh, F., Sabzalian, M., Christensen, J., “Phytoremediation of an aged petroleum contaminated soil using endophyte infected and non-infected grasses”, *Chemosphere,* Vol. 81, No. 9, 2010, pp. 1084–1090.
17. Kanaly, R.A. and Harayama, S., “Advances in the field of high-molecular-weight polycyclic aromatic hydrocarbon biodegradation by bacteria”, *Microb. Biotechnol.,* Vol. 3, No. 2, 2010, pp. 136–64.
18. Su Y.H. and Zhu Y.G., “Transport mechanisms for the uptake of organic compounds by rice (*Oryza sativa*) roots”, *Environ. Pollut.,* Vol. 148, No. 1, 2007, pp. 94–100.

19. Li, H., Sheng, G., Sheng, W.T., Xu, O.Y., "Uptake of trifluralin and lindane from water by ryegrass", *Chemosphere,* Vol. 48, 2002, pp. 335–341.
20. Wild, E., Dent, J., Tomas, G.O., Jones, K.C., "Direct observation of organic contaminant uptake, storage, and metabolism with plant roots", *Environ. Sci. Technol.,* Vol. 39, 2005, pp. 3695–3702.
21. Chiou, C.T., Sheng, G.Y., Manes, M., "A partition-limited model for the plant uptake of organic contaminants from soil and water", *Environ. Sci. Technol.,* Vol. 35, 2001, pp. 1437–1444.
22. Howsam, M., Jones, K.C., Ineson, P., "PAHs associated with the leaves of three dediduous tree species. II: uptake during a growing season", *Chemosphere,* Vol. 44, 2001, pp. 155–164.

# Changes in Soil Properties and Quality for a *Eucalyptus* Introduction Area: A Case Study in Lancang County, Yunnan Province

X. Q. ZHAO[1,*], Q. YI[1], N. DING[2] and J. S. XIA[1]
[1]*School of Resources Environment and Earth Science, Yunnan University, Kunming 650091, China*
[2]*Diyuan real estate consultative evaluation Co.,Ltd, Hefei 230001, Anhui, China*

**ABSTRACT:** Establishment of exotic species in the natural secondary forest and *Pinus kesiya* forest regions is common in Lancang County, southwestern Yunnan province, but their effects on soil properties and quality have not been fully studied. This research analyzed the effects of *Eucalyptus* plantation on soil physical and chemical properties, and evaluated the soil quality after *Eucalyptus* plantation replaced natural secondary evergreen broad-leaved forest and *Pinus kesiya* plantation. It was found that soil porosity, soil water content, soil pH, soil organic matter, exchangeable calcium, exchangeable magnesium and microelement decreased, soil bulk density increased and soil aeration permeability became very low after establishment of *Eucalyptus*. However, with forest litter retained, the contents of soil N, P and K had been rising in the case of applying fertilizer at the initial stage of *Eucalyptus* introduction. The soil quality indexes (*SQI*) under *Eucalyptus* plantation were lower than those under the natural secondary evergreen broadleaved forest and *Pinus kesiya* plantation. The SQI of soils in *Eucalyptus* plantation which replaced Pinus kesiya plantation lands (SQI = 0.208) was lower than that of soils in *Eucalyptus* plantation which replaced natural secondary evergreen broad-leaved forest lands (SQI = 0.269). The study results showed that replacement of natural secondary evergreen broad-leaved forest and *Pinus kesiya* plantation by *Eucalyptus* plantation has changed soil properties and lowered soil quality. The secondary evergreen broad-leaved forest vegetation that restored naturally is more favorable to maintaining and improving soil quality.

## INTRODUCTION

*EUCALYPTUS* is one of the most widely planted species of trees in the world, outside its native habitat in Australia and Southeast Asia [1-2]. Planting area of *Eucalyptus* has been more than $2 \times 10^6$ ha in China and now China is the third largest producer of *Eucalyptus* in the world with a 10% growth rate [3–4]. The area of *Eucalyptus* plantation in Yunnan Province is the fourth largest in China [5]. These *Eucalyptus* plantation intersperse with the remnant natural secondary forests to form a mosaic of plantation/natural secondary forest landscapes. The evergreen broad-leaved forest, dry land, shrub forest and *Pinus kesiya* forest had been largely replaced by *Eucalyptus* plantation, which amounted to more than 97% of the *Eucalyptus* introduced areas. Obviously, the *Eucalyptus* introduction has changed the land cover types in Yunnan [6], and the land cover change could produce direct and profound impact on soil quality [7–8].

To preventing soil degradation for maintaining the sustainability of land use, it is critical to analyze soil physical and chemical properties, evaluate soil quality and assess changes in soil properties and quality due to land use change [9]. Soil physicochemical property is one of the most key factors to measure soil quality [10]. Soil quality, introduced by Warkentin and Fletcher in 1977 [11], refers to the capacity of a specific kind of soil to function, within natural or managed ecosystem boundaries, to sustain plant and animal productivity, to maintain or enhance water and air quality, and to support human health and habitation [12–13]. As a complex functional entity, soil quality cannot be measured directly, but it can be inferred by measuring soil physical and chemical properties, which serve as quality indicators [14–15]. Soil properties that are responsive to the change in the land use dynamics dur-

---

*Author to whom correspondence should be addressed.
E-mail: zhaoxq748@sohu.com or xqzhao@ynu.edu.cn Tel:13888949695

1544-8053/14/04 107-11

ing a short-term period are considered as suitable soil quality indicators [16]. Different soil properties have different roles in maintaining soil quality [17]. It was neither reliable nor accurate to evaluate and compare the changes of soil quality through just using a single soil property [18]. Therefore, most researchers expressed soil quality in terms of physical and chemical properties, but few studies have combined those two aspects to evaluate soil quality synthetically were issued [19,63]. However, given that soils keep a unique balance among their physical and chemical characteristics, soil quality indices should incorporate both soil physical and chemical properties [20] for better representing soil quality [21–22]. Therefore, some researchers proposed to synthesize those soil physical and chemical properties to describe the changes of soil quality quantitatively [9,20–21,23–24]. In present, the integrated evaluation methods of soil quality include mainly multiple variable indicator-based Kriging method, soil quality dynamic method, soil quality comprehensive score method, relative soil quality evaluation method, and others [18,25–27]. The key step in the soil quality assessment is to select suitable soil quality indicators [28]. Soil quality indicators refer to measurable soil attributes that influence the capacity of a soil to support production [29] or other specified functions [30]. These attributes could be soil physical, chemical and/or biological properties [22,29,31]. The inclusion of different properties makes it possible to better reflect the complexity of the soil system [32].

As a biodiversity-rich area, Lancang County became the largest *Eucalyptus* introduction region in Yunnan Province since the early of the 21st century. After *Eucalyptus* forest replaced the natural secondary evergreen broad-leaved forest and *Pinus kesiya* forest, some studies on soil properties were mainly focused on the physicochemical properties [4, 33] and to our knowledge no study on soil quality in this region has been carried out. Zhao *et al.* showed that after the *Eucalyptus uraphylla* spp. forest replaced the secondary evergreen broad-leaved forest and *Pinus kesiya*, the soil water content, the total soil porosity and capillary porosity decreased, and soil non-capillary porosity and soil bulk density increased [34]. But Liu *et al.* studies suggested that the contents of soil nutrients in *Eucalyptus* plantation were higher than the background values. With comparison to the secondary forestry, the contents of total nitrogen and available phosphorus of *Eucalyptus* plantation were much lower [4]. The objectives of this study are to: (1) evaluate impact of land cover change from natural secondary evergreen broadleaf forests and *Pinus kesiya* forests to *Eucalyptus* plantation forests on soil physical and chemical properties, and (2) evaluate the changes of soil quality in response to the conversion of *Eucalyptus* plantation forests from natural secondary evergreen broad-leaved forests land and *Pinus kesiya* forests, and (3) identify advantages and disadvantages associated with introduction of *Eucalyptus* plantations instruction.

## MATERIALS AND METHODS

### Site Description

The study areas, is in Pu'er city, Lancang county (99°29′E-100°35′E, 22°01′N-23°16′N), southwest Yunnan Province, southwest China (Figure 1). It has a surface area of 8807 km$^2$ and elevation ranges from 580 m to 2516 m above sea level. The study area is in a subtropical montane monsoon climate with abundant rainfall and sunshine. Mean annual air temperature is 19.1°C, and the maximum and minimum temperatures are 37.2°C and –1.4°C, respectively. Mean annual precipitation is 1643 mm, annual mean relative humidity is 79%, and annual mean sunshine is 2098 h. The main soil types are latosolic red soils and red soils. Although this region has a high biodiversity, the monsoon evergreen broad-leaved forest is the dominant vegetation. In consequence of the distinct vertical climate variation, vegetation distribution also shows a distinct vertical pattern, which includes tropical seasonal rainforests, tropical monsoon evergreen broad-leaved forests, warm coniferous forests (such as *Pinus kesiya*), semi-humid evergreen broad-leaved forests, and mid-montane humid evergreen broad-leaved forests.

### Sites and Soil Sampling

Soil samples were taken from the depth of 0–20 cm in January 2010. According to the different types of land cover before *Eucalyptus* introduction, five survey sample areas were chosen, which are 7-year-old *Eucalyptus* forest in the first rotation. In order to avoid the influences caused by environmental factors and reduce the differences between sample areas, in the nearby of established *Eucalyptus* forest survey samples area, we chose two sites of 11-year-old natural secondary evergreen broad-leaved forests land and three sites of 9-year-old *Pinus kesiya* forest land as reference. These sites have consistent background conditions and the same habitat conditions (altitude, aspect, slope and

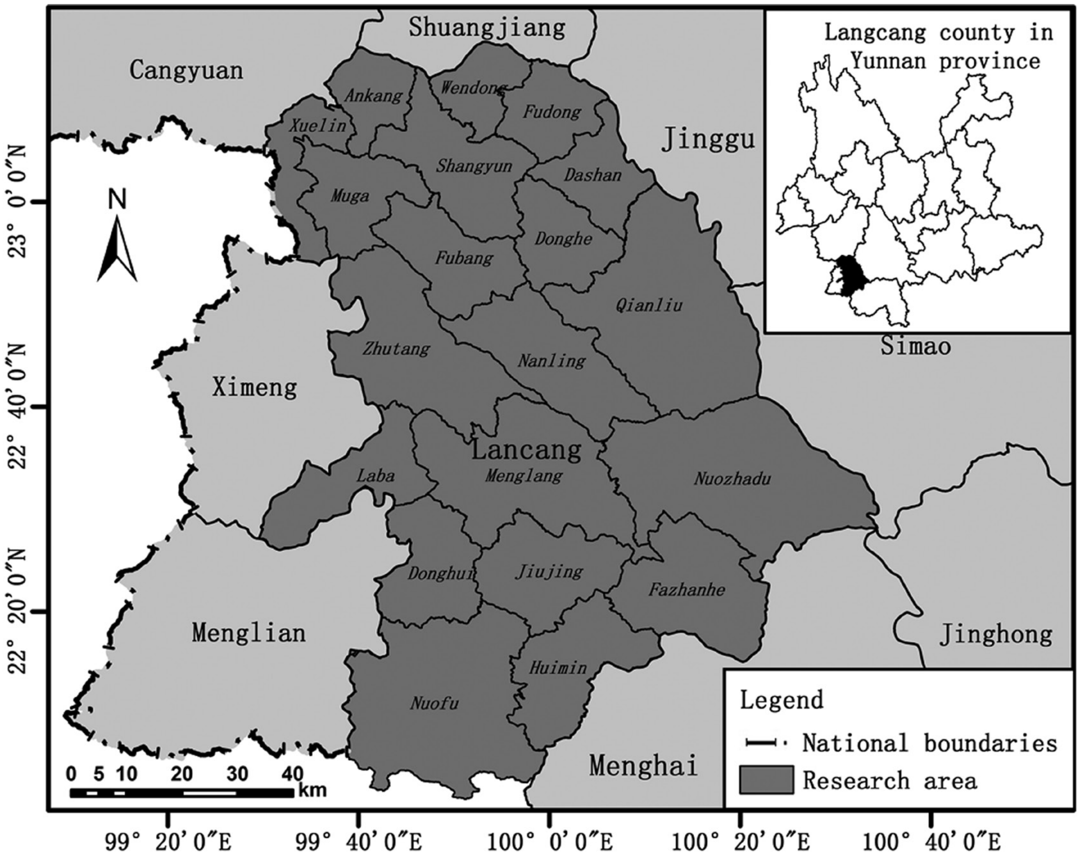


***Figure 1.*** *Location of the study site in the southwest Yunnan Province.*

edaphic characteristics) with *Eucalyptus* plantation forest sites. Three plots are deployed in each site with an equilateral triangle shape and about 50 m apart between each two plots. Soil samples were taken in five replicates from four corners and the center of a square plot of 20 m × 20 m area on each plot with hand trowel uniformly along same layer. 75 soil samples were collected on *Eucalyptus* forest land, 30 soil samples were collected on natural secondary evergreen broad-leaved forest land, 45 soil samples were collected on *Pinus kesiya* forest land. The collected soil samples from each pit and each replicate were mixed to form a bulk sample of about 1 kg, then were air-dried, mixed and passed through a 2 mm sieve for chemical analysis. Soil samples for bulk density and porosity determination were taken from the wall of the three pits using 100 $cm^3$ metal cylinder. Soil samples were taken from the field and stored in aluminum boxes for soil water content analysis.

## Analytical Methods

Soil bulk density and porosity were measured using the cutting ring method. The oven drying method was used to determine soil water content [35]. Soil pH value was measured using the potentiometer method [36] and soil organic carbon was determined by potassium dichromate oxidation [37]. Total nitrogen in soils was determined by Kjeldahl method [38], and hydrolysable nitrogen was determined using the standard method of forest soil hydrolytic nitrogen determination. Available phosphorus was determined by the double acid leaching molybdenum antimony colorimetric method [39]. Available K was determined by neutral leaching ammonium acetate solution, flame photometer method [40]. Available Fe, Zn, Mn, Cu were determined by Inductively Coupled Plasma Mass Spectrometry (ICP – MS).

To assess soil quality, we developed an integrated soil quality assessment method based on a combination of soil physical and chemical properties measured in the study area. Based on the assumption of different nutrients having different roles in maintaining soil quality, soil quality index (SQI) was developed and calculated through selecting soil quality indicators based on soil physical and chemical properties. Based on the correlation matrix and factor analysis, independent soil variables with higher component loadings or principal component eigenvalues greater than one were selected and included in the next step of the *SQI* assessment. The weighted average of these selected soil variables was obtained from the output of principal component analysis (PCA) and then the membership value *Q* (*Xi*) of each selected soil variable was determined. Finally obtained soil quality scores (*Wi*) and *Q* (*Xi*) were integrated to yield *SQI* as given in the following equation [24,41–42]:

$$SQI_j = \sum_{i=1}^{n} W_i \times Q(X_i) \qquad (1)$$

$$SQI = \sum_{j=1}^{m} SQI_j \times PCCR \quad (j = 1,2,\ldots,m) \qquad (2)$$

where $SQI_j$ is soil quality index of principal component *j*, *Wi* is the weighted vector ith soil quality factor and *Q*(*Xi*) is principal component value of each soil quality factor, *SQI* is the integrated soil quality index, *PCCR* is principal component contribution rate, *m* is number of principal component (PC). The values of *Q*(*Xi*) were calculated by ascending or descending functions using Equations (3) and (4), respectively [41].

$$Q(X_i) = (X_{ij} - X_{i\min})/(X_{i\max} - X_{i\min}) \qquad (3)$$

$$Q(X_i) = (X_{i\max} - X_{ij})/(X_{i\max} - X_{i\min}) \qquad (4)$$

where $X_{ij}$ is the value of the soil physical and chemical properties that were selected for the soil quality assessment, $X_{i\max}$ and $X_{i\min}$ are the maximum and minimum value of *i*th soil properties. If principal component factors are positive values, the factors were arranged in ascending order to compute *Q*(*Xi*) in Equation (3). If principal component factors are negative values, the factors were arranged in descending order to compute *Q*(*Xi*) using Equation (4).

### Statistical Analyses

One-way analysis of variance (ANOVA-1) was performed to assess the effect of land cover change on soil properties and quality using the MSTATC statistical software package. Partial correlation analysis was used to evaluate the correlation among variables. The data was statistically analyzed and the means were separated using the least significant difference at 0.05 or 0.01 probability levels using SPSS 14.0 software.

## RESULTS AND DISCUSSION

### Physical Properties of Soils Under the Natural Secondary Forest (NF), *Pinus kesiya* Forest (PF) and *Eucalyptus* Plantation Forest (EF)

Soil bulk density reflects the soil porosity status, air and water permeability, and the resistive state of root growth [43]. The larger soil bulk density is, the more compact the soils are, and the lower the air and water permeability are. Conversely, the smaller soil bulk density is, the less compact the soils are, and the permeability would be higher. But if the bulk density is too low, the soil could not support plant growth [44].

Soil bulk density (BD) in the *Eucalyptus* plantation forest land which replaced natural secondary evergreen broad-leaf lands (ENF), the *Eucalyptus* plantation land which replaced *Pinus kesiya* lands (EPF) and the PF land did not differ significantly except for the NF land ($P < 0.05$). The BD of NF was 1.016 g/cm$^3$, less than those of PF and EF (Table 1), and the order of BD value is EPF > PF > ENF > NF. After EF replaced NF and PF, soil bulk density increased by 17.72% and 7.91% respectively in the first rotation of the cutting

***Table 1. Comparison of Physical Properties Under Different Land Cover Types.***

| | BD/g·cm$^3$ | | TP/% | | CP/% | | NCP/% | | SWC/% | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Land Cover Types** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** |
| ENF[a] | 1.034 | 0.37 | 59.85 | 0.734 | 43.93 | 0.374 | 17.04 | 0.74 | 22.71 | 0.854 |
| NF[b] | 1.016 | 0 | 62.75 | 0.032 | 45.4 | 0 | 16.23 | 0.08 | 24.62 | 0.496 |
| EPF[c] | 1.405 | 0.7 | 48.98 | 0932 | 40.9 | 0.696 | 7.08 | 0.87 | 21.61 | 0.554 |
| PF[d] | 1.302 | 0.67 | 50.87 | 0.993 | 43.6 | 0.666 | 6.26 | 0.27 | 23.08 | 0.391 |

*Notes:*

[a] *Eucalyptus* plantation which replaced natural secondary evergreen broad-leaf forest.

[b] Natural secondary evergreen broad-leaf forest.

[c] *Eucalyptus* plantation which replaced *Pinus kesiya* forest.

[d] *Pinus kesiya* forest; BD: bulk density; TP: total porosity; CP: capillary porosity; NCP: non-capillary porosity; SWC: soil water content.

***Table 2. Comparison of Chemical Properties Under Different Land Cover Types.***

| | pH | | SOM/g.kg$^{-1}$ | | TN/% | | HN/% | | AP/mg.kg$^{-1}$ | | AK/mg.kg$^{-1}$ | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Land Cover Types** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** |
| ENF | 5.36 | 0.898 | 42.7 | 0.192 | 21.01 | 0.34 | 160.61 | 0.018 | 5.18 | 0.021 | 62.75 | 0.016 |
| NF | 5.38 | 0.058 | 45.9 | 0.262 | 20.04 | 0 | 148.18 | 0 | 3.71 | 0.005 | 48.61 | 0.001 |
| EPF | 5.22 | 0.877 | 18.39 | 0.155 | 10.11 | 0.339 | 82.02 | 0.83 | 2.64 | 0.049 | 101.9 | 0.216 |
| PF | 5.34 | 0.325 | 20.67 | 0.104 | 7.72 | 0.018 | 54.72 | 0.006 | 1.96 | 0.001 | 80.46 | 0.406 |

*Note:* pH: soil pH; SOM: soil organic matter; TN: total nitrogen; HN: hydrolysable nitrogen; AP: available phosphorus; AK: available K; For ENF, NF, EPF, PF abbreviations see Table 1.

period, which is consistent with some previous studies [45–47]. Those studies all showed that introduction of EF had changed the physical structure of soils and increased soil compactness.

Soil porosity is an important indicator of soil structure. The larger soil porosity is, the less compact soil structure is, which would be favorable for the rapid infiltration of rainfall and the reduction of surface runoff scouring [48–50].

Total soil porosity (TP) and soil capillary porosity (CP) of the EF and PF did not differ significantly except for the NF ($P < 0.05$). soil non-capillary porosity (NCP) did all not differ significantly ($P > 0.05$). The order of soil TP and CP were the same, i.e., NF > ENF > PF > EPF and the order of soil NCP was ENF > NF > EPF > PF. After EF replaced NF and PF, soil total porosity decreased by 4.62% and 3.72%, respectively. Capillary porosity also decreased by 3.24% and 6.19%, respectively. However, non-capillary porosity increased by 4.99% and 13.10% (Table 1). Table 1 also showed that compared with EF, NF and PF possessed less compact soils and with higher permeability, and such soils with large storage capacity and high conductivity of water are favorable for vegetation growth. However, the EF is associated with high interception capacity that can reduce surface runoff and thus decrease the potential of soil scouring and erosion. Guo *et al.* also found that soil total porosity, capillary porosity and non-capillary porosity decreased after *Eucalyptus* plantation replaced natural secondary forest [46].

Soil water is a requirement and a participant as well for many physical, chemical and biological processes, while it acts as the main input channels of nutrients for plants [51]. Soil water content (SWC) of the EF, NF and PF did all not differ significantly ($P > 0.05$) but the soil water contents under these three land covers were slightly different. Soil water content of NF was the highest, PF came second, and EF's was the lowest, and the order of SWC was NF > PF > ENF > EPF. After EF replaced NF and PF, soil water content decreased by 7.76% and 6.37% respectively (Table 1), which indicates that the soil water consumption in the EF was the largest. The study results were in agreement with some scholars' findings [53], but were opposite to other scholars' findings[46].

In conclusion, *Eucalyptus* plantation introduction changed soil physical properties, i.e., soil water content, soil total porosity and capillary porosity decreased, while soil bulk density and soil non-capillary porosity increased. After EF replaced NF and PF, soil physical structure changed and the compactness and permeability of soils were affected, e.g., the compactness of soils increased, soil water retention capacity decreased, and soil water content reduced.

## Chemical Properties of Soils under the NF, PF, and EF

The soil total nitrogen (TN) and hydrolysable nitrogen (HN) of the EF did not differ significantly except for the NF and PF ($P < 0.05$). The soil TN and HN contents of ENF were the largest among ENG, NF, EPF and PF, which were 21.01% and 160.61%, respectively (Table 2). The soil TN and HN contents of PF were the lowest, which were 7.72% and 54.72%, respectively. Table 2 showed that soil TN and HN contents of EF were larger than those of NF and PF. The orders of soil TN and HN contents were the same, which were ENF > NF > EPF > PF. After EF replaced NF and PF, soil TN and HN contents increased by 4.84%, 30.96%, and 8.39%, 49.89%, respectively.

Soil available phosphorus (AP) of the EF did not differ significantly except for the NF and PF ($P < 0.05$). Just as it was for soil N content, the soil AP content of EF which replaced NF was the largest, which was 5.18 mg/kg. Soil AP content of PF was the lowest, which was 1.96 mg/kg (Table 2), and the order of soil AP content was ENF > NF > EPF > PF. It showed that soil AP content of EF was higher than that of NF and PF. After NF and PF are replaced by EF, the soil AP contents are increased by 39.62% and 34.69%, respectively.

Soil available K of the PF and the EF which replaced the PF did not differ significantly except for the NF and EF which replaced NF ($P < 0.05$). The soil available K content of the EPF was the highest, and its content was 101.9 mg/kg. Soil available K of the NF was the lowest, and its content was 48.61 mg/kg (Table 2), and the order of soil available K was EPF > PF > ENF > NF. Table 2 showed that soil available K content of the EF was higher than that of the NF and PF. After the NF and PF are replaced by EF, the soil available K content is increased by 29.09% and 26.65%, respectively.

Applying N, P, K fertilizers at the primary stage of *Eucalyptus* introduction, the contents of soil N, P, K increased after *Eucalyptus* plantation replaced natural secondary evergreen broad-leaved forest and *Pinus kesiya* forest during the first cutting period of *Eucalyptus*, and this was similar to some previous study results [53–54]. But some studies [47,55–56] found that the contents of soil N, P, K decreased after *Eucalyptus* plantation replaced other tree species, and the contents of soil N, P, K became very low. The reason is that the soil properties vary greatly in different regions. Simultaneously, during the process of *Eucalyptus* plantation introduction, different management and fertilization practices would have great influences on soil N, P, K contents.

## Soil pH, Organic Matter

Soil pH of the EF, NF and PF did all not differ significantly ($P > 0.05$). Soil pH value of the NF was 5.38, which was larger than the pH values of the PF and EF (Table 2), and the order of pH values was NF > ENF > PF > EPF. After the NF and PF are replaced by the EF, soil pH is decreased by 0.37% and 2.25%, respectively, which indicates that the introduction of EF can increase acidity of the soils.

Soil organic matter (SOM) of the EF, PF and NF did all not differ significantly ($P > 0.05$). The SOM of the NF was 45.9 g/kg, which was larger than the value of the PF and EF (Table 2), and the order of SOM values was NF > ENF > PF > EPF. After the NF and PF are replaced by the EF, the SOM is decreased by 6.97% and 11.03%, respectively, which indicates that the SOM consumption by the EF is higher than the other species.

Soil microelements directly impact the growth of vegetation. Sometimes they are even more important than major elements, because microelements in soils reflect the level of soil supply for the plant with mineral nutrients, and therefore the concentration of soil microelement is an important indicator of soil quality [57]. Soil microelements including soil available Fe, Zn, Cu, Mn, B in the EF, PF and NF did not differ significantly ($P > 0.05$) (Table 3).The orders of soil available Fe, Zn, Cu were the same, i.e., PF > EPF > NF > ENF, but the order of soil available Mn was NF>EFN>PF>EFP, and the order of soil available B was NF > PF > ENF > EPF. After the EF replaced the NF and PF, soil available Fe, Zn, Cu, Mn and B decreased by 7.06%, 11.14%, 9.39%, 25.58%, 19.62% and 3.42%, 10.07%, 20.74%, 7.34%, 37.56% respectively, which indicates that the soil microelement consumption by EF was bigger than other species.

After *Eucalyptus* plantation replaced natural secondary evergreen broad-leaved forest and *Pinus kesiya* forest, the pH value, soil organic matter and microelement all decreased. This result is consistent with the majority of previous study results. For example, Gong and Liao found that soil organic matter content under *Eucalyptus* plantation was significantly lower than those under the natural forests, and which would result in lower soil cation exchange capacity [47]. Huang and Yu studied the different contents of soil organic matter under 6 species of *Eucalyptus* trees and *Pinus yunnanensis* in Fumin County of Yunnan Province, and found that soil organic matters under *Eucalyptus* trees were significantly lower than those under *Pinus yunnanensis* [56]. By studying soil chemical properties

***Table 3. Comparison of Microelement Under Different Land Cover Types.***

| | Fe/g.kg$^{-1}$ | | Zn/g.kg$^{-1}$ | | Cu/g.kg$^{-1}$ | | Mn/g.kg$^{-1}$ | | B/g.kg$^{-1}$ | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Land Cover Types** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** | **Mean** | **Sig.** |
| ENF | 29.399 | 0.084 | 0.197 | 0.729 | 0.016 | 0.559 | 0.222 | 0.983 | 0.878 | 0.297 |
| NF | 31.634 | 0.822 | 0.222 | 0.777 | 0.018 | 0.671 | 0.299 | 0.894 | 1.092 | 0.308 |
| EPF | 40.577 | 0.147 | 0.257 | 0.284 | 0.019 | 0.809 | 0.201 | 0.706 | 0.572 | 0.773 |
| PF | 42.013 | 0.282 | 0.286 | 0.854 | 0.024 | 0.784 | 0.217 | 0.493 | 0.917 | 0.393 |

*Note:* Fe, Zn, Cu, Mn, B: available Fe, Zn, Cu, Mn, B; For ENF, NF, EPF, PF abbreviations see Table 1.

**Table 4. The Correlations Between SQI and Each Evaluation Indicator.**

| SQI | BD | TP | CP | NCP | SWC | pH | SOM | TN | HN |
|---|---|---|---|---|---|---|---|---|---|
| 1 | **–0.870**** | **–0.650*** | **0.810**** | **0.785**** | 0.456 | 0.066 | **0.897**** | **0.814**** | **0.732**** |

| SQI | AP | AK | Fe | Zn | Cu | Mn | B |
|---|---|---|---|---|---|---|---|
| 1 | **0.597*** | – 0.293 | **–0.807**** | **–0.807**** | **–0.849**** | **0.808**** | 0.119 |

*Note:* For abbreviations see Table 1, 2 and 3.
*Indicates significant differences at $p < 0.05$; **Indicates significant differences at $p < 0.01$.

under *Eucalyptus* forest in Leizhou Peninsula, Zhong *et al.* showed that soil microelements of available Cu, Zn, Mn, B were very low. However, by studying soil microelements of B, Cu, Zn, Fe, Mn in evergreen broad-leaved forest, *Pinus kesiya* forest and 3-year-old *Eucalyptus* plantation in the southwest of Yunnan [58], Yuan, Zhao and Yang found that other soil microelements contents in *Eucalyptus* forest were larger than the contents of the evergreen broad-leaved forest and the *Pinus kesiya* forest except for soil available Fe and Zn. This may be related to the young age of *Eucalyptus* plantation. With the increasing of age, the soil microelements consumption in *Eucalyptus* plantation will increase [59–60].

## Soil Quality Integrated Index Under the NF, PF, and EF

### *Selection of Evaluation Factors in the Integrated Soil Quality Index*

Significant correlation between soil properties is the precondition of screening main evaluation indicators of soil quality. According to the above 16 indicators and Equations (1)–(5), SQI was calculated. By analyzing the correlations between 16 evaluation indicators and SQI (Table 4), at the 0.01 confidence level, SQI had obvious positive correlations with soil capillary porosity, non-capillary porosity, soil organic matter, soil total nitrogen, hydrolyzed nitrogen and available Mn, and it had obvious negative correlations with soil bulk density, available Fe, available Zn, available Cu. At the 0.05 confidence level, SQI had obvious positive correlations with available phosphorus, and it had obvious negative correlations with total porosity.

A regression analysis of soil bulk density and soil porosity yielded the following expression:

$$BD = 2.650 - 1.337\mathrm{E} - 9CP - 2.650TP - 1.064NCP$$

where $BD$ was the soil bulk density, $CP$ was capillary porosity, $TP$ was total porosity, $NCP$ was non capillary porosity. At $P = 0.000$, $R = 1.000$, $Ra = 1.000$, the results showed that soil bulk density had strong correlations with soil total porosity, capillary porosity and non-capillary porosity. Consequently, the soil bulk density indicator could be used to replace soil porosity indicator in evaluating SQI.

Thus, nine indicators including soil bulk density, soil organic matter, soil total nitrogen, hydrolyzed nitrogen, available phosphorus, available Fe, available Zn, available Mn and available Cu were selected as evaluation indicators of soil quality.

### *Evaluation of the Integrated Soil Quality Index*

Firstly, a factor analysis was performed among

**Table 5. The Principal Component Eigenvalues and Contribution Rate of Soil Quality Factors.**

| Principal Components | Eigenvalues | Contribution Rate, % | Accumulating Contribution Rate, % |
|---|---|---|---|
| 1 | 3.919 | 43.547 | 43.547 |
| 2 | 2.279 | 35.326 | 78.873 |
| 3 | 0.784 | 6.712 | 85.585 |
| 4 | 0.707 | 5.855 | 91.440 |
| 5 | 0.527 | 4.855 | 96.295 |
| 6 | 0.317 | 2.018 | 98.313 |
| 7 | 0.2 | 1.219 | 99.532 |
| 8 | 0.177 | 0.466 | 99.998 |
| 9 | 0.09 | 0.002 | 100 |

*Table 6. The Load and Weight of Soil Quality Factors on Each Principal Component.*

| Soil Quality Factors | PCL1 | Weight Value 1 | PCL2 | Weight Value 2 |
|---|---|---|---|---|
| BD | –0.711 | 0.135 | 0.131 | 0.035 |
| SOM | 0.876 | 0.166 | 0.321 | 0.086 |
| TN | 0.893 | 0.169 | 0.244 | 0.065 |
| HN | 0.858 | 0.163 | 0.108 | 0.029 |
| AP | 0.817 | 0.155 | 0.057 | 0.015 |
| Fe | –0.590 | 0.122 | 0.713 | 0.191 |
| Zn | –0.125 | 0.024 | 0.605 | 0.162 |
| Cu | –0.225 | 0.043 | 0.798 | 0.214 |
| Mn | 0.173 | 0.033 | 0.757 | 0.203 |

*Note:* BD: Soil bulk density; SOM: Soil organic matter; TN: Total soil N; HN: Hydrolysable nitrogen; AP: Soil available phosphorus; Fe, Zn, Cu, Mn, Soil available: Fe, Zn, Cu, Mn, PCL1 and PCL2: The first principal component load and the second principal component load.

the nine indicators of soil quality. The iterated PCA showed that the first two PCs with eigenvalues greater than one explained 78.87% of the variability, and could adequately represent the information contained in the original variable (Table 5). Table 6 shows the correlation degree between each principal component (common factor) and original factor, where the plus sign and minus sign respectively represent the positive correlation and negative correlation. The greater the absolute value is, the higher the correlation degree is. The loadings of soil bulk density, available Fe, available Zn, available Cu were negative in the first principal components, and the others were positive; All factor loadings were positive in the second principal components. Equations (3) and (4) were used to determine membership values $Q(Xi)$ of different soil quality factors (Table7). The weight coefficients ($Wi$) of these two principal components were calculated according to their loadings. The weights of the indicators $Wi$ and the membership values $Q(Xi)$ used to assess *SQI*s can reflect the relative soil quality of different land covers, which are shown in Table 6 and Table7 for the selected soil properties.

Equations (1) and (2) were used to calculate the integrated soil quality index (SQI) of different land covers (Figure 2). The higher *SQI* value is, the better soil quality is, and vice versa. The *SQI* was 0.323 for the NF, 0.228 for the PF, 0.269 for the ENF, and 0.208 for the EPF, which indicates that the land cover/land use plays an important role in affecting soil quality. After the EF replaced the NF and PF, the integrated soil quality index decreased by 20.07% and 9.64%, which showed that *Eucalyptus* introduction degraded soil quality. Especially after the NF was replaced by the EF, the decline rate of the soil quality was higher, but in the place where the EF replaced PF, the decline rate was comparatively less.

With different ways of land use and management, various land covers resulted in differences in soil nutrient enrichment and redistribution effect [61–63], and which would have an impact on the soil physicochemical properties and soil quality [64–65]. Some studies suggested that *Eucalyptus* plantation introduction could consume a large amount of soil fertility and

*Table 7. The Membership Values of Soil Quality Factors Under Different Land Cover Types.*

| | ENF | | NF | | EPF | | PF | |
|---|---|---|---|---|---|---|---|---|
| Soil Quality Factors | PCM1 | PCM2 | PCM1 | PCM2 | PCM1 | PCM2 | PCM1 | PCM2 |
| BD | 0.532 | 0.468 | 0.658 | 0.342 | 0.329 | 0.671 | 0.477 | 0.523 |
| SOM | 0.349 | 0.349 | 0.411 | 0.411 | 0.208 | 0.208 | 0.205 | 0.205 |
| TN | 0.336 | 0.336 | 0.540 | 0.540 | 0.181 | 0.181 | 0.208 | 0.208 |
| HN | 0.362 | 0.362 | 0.461 | 0.461 | 0.159 | 0.159 | 0.224 | 0.224 |
| AP | 0.233 | 0.233 | 0.351 | 0.351 | 0.214 | 0.214 | 0.353 | 0.353 |
| Fe | 0.539 | 0.461 | 0.520 | 0.480 | 0.434 | 0.566 | 0.480 | 0.520 |
| Zn | 0.652 | 0.348 | 0.666 | 0.334 | 0.739 | 0.261 | 0.712 | 0.288 |
| Cu | 0.509 | 0.491 | 0.538 | 0.462 | 0.474 | 0.526 | 0.771 | 0.229 |
| Mn | 0.308 | 0.692 | 0.488 | 0.512 | 0.225 | 0.775 | 0.306 | 0.694 |

*Note:* For abbreviations see above table.

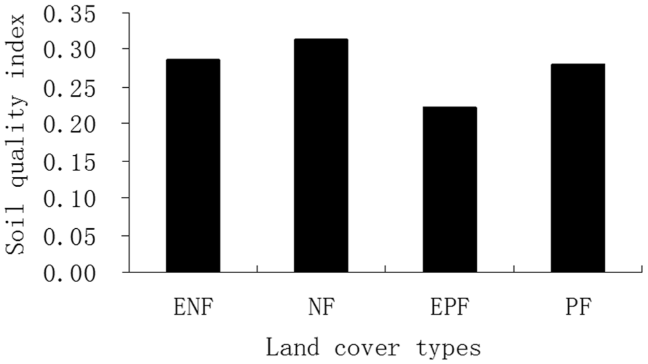


***Figure 1.*** *Soil quality index (SQI) under different land cover types. Note: For abbreviations see above table.*

water, leading to the soil quality degradation [66–67]. Whereas, some researchers suggested that Eucalyptus plantation could improve the environment, prevent soil and water loss, and maintain soil quality under the condition of non-intensive management [68]. Zhong et al. evaluated comprehensively soil fertility of Masson pine, *Acacia mangium* and *eucalypt* using the improved Nemerow comprehensive index method and found that eucalypt forest had less effect on soil nutrient [69].

In this study, the SQI of soils under the EF were lower than those under the NF and PF, which may be due to the fast-growing nature of the *Eucalyptus* species that may intensively absorb soil nutrients. This result is consistent with some previous research results [66]. After the EF replaced the NF, soil quality decreased greatly. One possible reason is that the soils in the NF had higher species richness and better micro ecological environment (such as light, soil moisture, surface temperature, biodiversity, microbial biomass and activity, and etc.), which helped to maintain high soil quality [22]. However, after the EF replaced the PF, soil quality declined at a relatively slow rate. On the one hand, this could be attributed to the short time period of *Eucalyptus* introduction (i.e., seven years), and the changes of soil original properties were relatively small in such short time period; on the other hand, the relevant management measures involved in the *Eucalyptus* introduction were more effective, such as spreading fertilizer and keeping the litters at the early stage of the EF introduction. Nevertheless, even for the EF, soil qualities were of obvious differentials due to the distinctions of previous land use types. Soils under the EF that replaced the PF (SQI = 0.208) had lower values of SQI compared with those established on the NF (SQI = 0.269). This result demonstrates the extent to which soils under different land cover types differ in quality.

## CONCLUSIONS

Land cover types change from natural second green broad-leaved forest and *Pinus kesiya* plantation to *Eucalyptus* plantation decreased soil porosity, soil water content, soil pH, soil organic matter, exchangeable calcium, exchangeable magnesium and microelement, increased soil bulk density, and lowered soil aeration permeability. Because fertilization and good management measures, the contents of soil N, P, K increased at the initial stage of *Eucalyptus* introduction. Soil quality of the natural secondary evergreen broad-leaved forest was the best, but after it was replaced by the *Eucalyptus* plantation, the soil quality decreased greatly. By comparison, where *Pinus kesiya* forest was replaced by *Eucalyptus* plantation, soil quality declined slightly. It suggests that the secondary evergreen broad-leaved forest vegetation of natural recovery was more favorable to maintaining and improving soil quality, and reasonable management measures play a very important role for maintaining the soil quality in the *Eucalyptus* introducing areas.

## ACKNOWLEDGEMENTS

The research was funded by project of the National Natural Science Foundation of China (Grant No. 41361020, 40961031, 41461103).

## REFERENCES

1. Westoby J. C. (1989): *Introduction to World Forestry.* Blackwells. Oxford University Press, Oxford, UK, 228.
2. Evans J. (1992): *Plantation Forestry in the Tropics. 2nd edn.* Oxford University Press, Oxford, UK, 403.
3. Huang Y. Y., Zhong X. Q., Wang J. Z., Ni C. Y., Li Q. X. (2007): The eco-economics problems which China should attend in the process development *Eucalyptus* paper making. *Ecological Economy,* 8:38–41.
4. Liu P., Qin J., Liu J. C., Li Y. Z., Wang H. F., Wang X. K. (2011): Analysis and evaluation of soil nutrients and heavy metals of *Eucalyptus* plantation. *Chinese Journal of Environmental Engineering, 5*(3):649–656 (In Chinese).
5. Zhang R. G., Li S. G., Jiang Y. D. (2007): Introduction of *Eucalyptus* in Yunnan and Analysis on Development Status. *West China Forestry Science, 36*(3):97–102 (In Chinese).
6. Ding N. (2013): Study on the changes in soil quality after *Eucalyptus* introduction in Lancang county:[Dissertation]. Kunming: Yunnan University (In Chinese).
7. Zhu Z. X. (1983): Soil science. China, Beijing: China agriculture press, 276–279 (In Chinese).
8. Wu J. G. Zhang X. Q. Xu D. Y. (2004): Impact of land-use change on soil carbon storage. *Chinese Journal of Applied Ecology, 15*(4): 593–599 (In Chinese).
9. Liu S. L., Fu B. J., Chen L. D., Qiu J., Lu Y. H. (2003): Comparison of two quantitative methods in assessing soil quality in different land uses. *Resources and Environment in the Yangtze Basin, 12*(5):422–426 (In Chinese).

10. Ding S. L., Yang N. G., Zhao C.C., Yang Q. M., Wang S., Dong X. (2010): Soil Physical and Chemical Properties in Water Conservation Forest in Eastern Qinghai Province. *Bulletin of Soil and Water Conservation, 30*(6):1–6 (In Chinese).
11. Wienhold B. J., Andrews S. S., Karlen D. L. (2005): Soil quality: indices and appraisal. *Indian: Indian Society of Soil Science,* 67–72.
12. Doran J. W., Parkin J. B. (1994): Defining and assessing soil quality. In: J. W. Doran, D. C. Coleman, D. F. Bezdicek, & B. A. Stewart (Eds.), *Defining soil quality for sustainable environment,* (35):3–21. Soil Science Society of America (SSSA) Special Publication No. 35. Madison, WI: ASA and SSSA.
13. Islam K. R., Weil R. R. (2000): Soil quality indicator properties in mid-Atlantic soils as influenced by conservation management. *Journal of Soil And Water Conservation, 55*: 69–78.
14. Brejda J. J., Moorman T. B. (1999): Identification and interpretation of regional soil quality factors for the central high plains of the midwestern USA. Sustaining the global farm. Selected papers from the 10th International Soil Conservation Organization Meeting held May 24–29.
15. Diack M., Stott D. E. (2001): Development of soil quality index for the Chalmers silty clay loam from the Midwest USA, In D. E. Stott, R. H. Mohtar, & G. C. Steinhardt (eds.), Sustaining the global farm: 550–555. Selected papers from the 10th international soil conservation organization meeting held on May 24–29, 1999 at Purdue University and the USDA-ARS National Soil Erosion Laboratory.
16. Carter M. R., Gregorich E. G., Angers D. A., Donald R. G., Bolinder M. A. (1998): Organic C and N Storage and Organic C Fractions in Adjacent Cultivated and Forested Soils of Eastern Canada. *Soil & Tillage Research, 47*:253–261.
17. Ambachew D., Bal R. S., Rattan L., Trond B. (2012): Effects of *Eucalyptus* and coniferous plantations on soil properties in Gambo District, southern Ethiopia. *Acta Agriculturae Scandinavica Section B—Soil and Plant Science, 62*: 455–466.
18. Wang X. J., Gong Z. T. (1998): Assessment and analysis of soil quality changes after eleven years of reclamation in subtropical China. *Geoderma, 81*:339–355.
19. Hobbs R. J., Huenneke L. F. (1992): Disturbance, diversity, and invasion: Implications for conservation. *Biological Conservation, 6*:324–327.
20. Barrios E., Delve R. J., Bejunda M., Mowo J., Agunda J., Ramisch J., Trejo M. T., Thomas R. J. (2006): Indicators of soil quality: a south–south development of a methodological guide for linking local and technical knowledge. *Geoderma 135*:248–259.
21. Dumanski J., Pieri C. (2000) : Land quality indicators: research plan. Agriculture, *Ecosystems & Environment, 81*:93–102.
22. Li L. (2010): Study on soil ecological effects of different land use in red soil hilly region of sourthern China:[Dissertation]. China, Kunming: Yunnan University, 06:31–32 (In Chinese).
23. Adejuwon J. O., Ekanade O. (1988): A comparison of soil properties under different land use types in a part of the Nigerian cocoa belt. *Catena, 15*: 319–331.
24. Zhang Q. F. Song Y. C. You W. H. (1999): Relationship between plant community secondary succession and soil fertility in TianTong, ZheJiang Province, *Acta Ecologica Sinica, 19*(2): 174–178 (In Chinese).
25. Smith J. L., Halvorson J. J. (1993): Papendick R I. Using multiple-variableindicator kriging for evaluating soil quality. *Soil Science Society of America Journal, 57*: 743–749.
26. Kwansoo Kim, Bradford L. Barham, Ian Coxhead (2001): Measuring soil quality dynamics: A role for economists, and implications for economic analysis. *Agricultural Economics, 25*: 13–26.27.
27. Cambardella C. A ., Moorman T. B., Andrews S. S., *et al.* (2004): Watershed-scale assessment of soil quality in the loess hills of southwest Iowa. *Soil & Tillage Research, 78*: 237–247.
28. Liu Z. F. Fu B. J. Liu G. H., *et al.* (2006): Soil quality: Concept indicators and its assessment *Acta Ecologica Sinica, 26*( 3) : 901–13 ( in Chinese).
29. Arshad M. A., Martin S. (2002): Identifying critical limits for soil quality indicators in agro-ecosystem. *Agriculture, Ecosystems & Environment, 88*:153–160.
30. Karlen D. L., Mausabach M. J., Doran J. W., Cline R. G., Harris R. F., Schuman G. E. (1997): Soil quality: a concept, definition and framework for evaluation. *Soil Science Society, 61*:4–10.
31. Nambiar K. K. M., Gupta A. P., Fu Q., Li S. (2001): Biophysical, chemical and socio-economic indicators for assessing agricultural sustainability in the Chinese coastal zone. *Agriculture, Ecosystems & Environment, 87*:209–214.
32. Zornoza R., Mataix-Solera J., Guerrero C., Arcenegui V. A., Mayoral M., Morales J., Mataix-Beneyto J. (2007): Soil properties under natural forest in the Alicante Province of Spain. *Geoderma, 142*:334–341.
33. Zhao X. Q., He C. L., Xu X. H. (2012): The effect of *Eucalyptus uraphylla spp.* Introduction on soil physical properties in a mountainous region of Yunnan. *Ecology and Environmental Sciences, 21*(11):1810–1816 (In Chinese).
34. Zhao X. Q., Ding N., Yan P. (2012): Changing Rules of Physical and Chemical Properties of *Eucalyptus* uraphylla spp. Forest at Different Ages in Southwest Yunnan Province. *Agricultural Science & Technology, 13*(6): 1298–1302(In Chinese).
35. Forestry department Science and Technology Division (1994): *Forest ecosystem positioning methods.* China, Beijing: China Science and Technology Press, 98–100 (In Chinese).
36. Kim H. T. (2005): *Soil sampling, preparation, and analysis (2nd Edn.)*. London: Taylor & Francis.
37. Nelson D. V., Sommers L. E. (1982): Total carbon, organic carbon, and organic matter. In: Page, A.L. (Ed.), Methods of soil analysis, Part 2. Soil Science of America, Madison, Wisconsin.
38. Bremner J. M., Mulvaney C. S. (1982): Nitrogen Total. Part II. In: "*Methods of Soil Analysis*", (Ed.): Bu Buxton, D. R.. ASA, Monograph, No. 9, 2nd Edition, Madison, WI, 595–624.
39. Nanjing Institute of Soil Science, CAS (1981): *Analysis of soil physical and chemical properties.* China, Shanghai: Shanghai Sci. & Tech. Press.
40. Black C. A., Evans D. D., White J. L., Ensminger L. E., Clark F. E. (1965): *Methods of Soil Analysis. Part 1. Physical and Mineralogical Properties, Including Statistics of Measurement and Sampling.* Am. Soc. Agro. Inc., Madison, WI.
41. Fu B. J., Liu S. L., Chen L. D., Lu Y. H., Qiu J. (2004): Soil quality regime in relation to land cover and slope position across a highly modified slope landscape. *Ecological Research, 19*:111–118.
42. Masto R. E., Chhonkar P. K., Singh D., Patra A. K. (2007): Soil quality response to long-term nutrient and crop management on a semi-arid *Inceptisol. Agriculture, Ecosystems and Environment, 118*:130–142.
43. Liu S. L., Fu B. J., Ma K. M., Liu G. H. (2004): Effects of vegetation types and landscape features on soil properties at the plateau in the upper reaches of Minjiang River. *Chinese Journal of Applied Ecology, 15*(l):26–30 (In Chinese).
44. Han Y. S., Wei Y. C., Ouyang Z. Y., Cao Y. (2008): Effects of continuous planting rotation on forest structural characteristics and water holding capacity of *Eucalyptus* plantations. *Acta Ecologica Sinica, 28*(9): 4609–4617 (In Chinese).
45. Xu L. B., Lu M. X., Shi M. (2007): Study on soil physical properties under *Eucalyptus* forest in west of Yunnan. *Journal of Shandong Forestry Science and Technology, 173* (6): 41–43 (In Chinese).
46. Guo J. Y., Shi Z. J., Xu D. P., Tai L. K., Zhou W. W., Yang Z. J. (2008): Effect of natural secondary forest and its artificial regeneration forests on soil physical properties and carbon storage in Hainan island, China. *Ecology and Environment, 17*(6): 2366–2369. (In Chinese).
47. Gong S. S., Liao S. G. (2009): Soil nutrient characteristics in eucalypt plantation and natural forest. *Journal of Jiangsu Forestry Science & Technology, 36*(3):1–4 (In Chinese).
48. Huang J. Y., Yan L. J., Wang Z. Q. (2002): The characteristics of soil and water loss of different land utilization models in small watersheds of red soil region. *Journal of Zhejiang University (Agric. & Life Sci.), 28(*1):78–82 (In Chinese).
49. Shi D. M., Lv G., Jiang G. Y., Lu X. P. (2005): Study on Anti-erodibility and Change of Soil Physical Property in *Pinus massoniana* Woodland. *Journal of Soil and Water Conservation,19*(6):35–39 (In Chinese).
50. Kang B., Liu S. R., Cai D. X., etal.( 2010): Soil physical and chemical characteristics under different vegetation restoration patterns in

China south subtropical area. *Chinese Journal of Applied Ecology, 21*(10):2479–2486 (In Chinese).

51. Chen A. L., Hong W. (2006): The Effects of Gaps in Central Subtropical Evergreen Broad-leaved Forest on Soil Fertility in Fujian Province. *Acta Agriculture Universitatis Jiangxiensis, 28*(5):723–727 (In Chinese).
52. Li Y. X., Luo W., Bei M. R. (2007): Influence of *Eucalyptus* Plantation on Soil Quality. *Journal of West China Forestry Science, 36*(4):100–104 (In Chinese).
53. Adrian Ares, James H. Fownes (2000): Productivity, nutrient and water-use effciency of *Eucalyptsus saligna* and *Toonaciliata* in Hawaii. *Forest Ecology and Management, 13*(9):227–236.
54. Jeddi K., Cortina J., Chaieb M. (2009): Acacia salicina, Pinus halepensis and *Eucalyptus* occidentalis improve soil surface conditions in arid southern Tunisia. *Journal of Arid Environments, 73*: 1005–1013.
55. Liao S., Ye Z. ,Wang Y. M., Feng H. H. (2008): A Comparative Study on Physical and Chemical Properties of Soils in *Eucalyptus* Plantations, Chinese Fir Plantations and Bamboo Forest. *Journal of Subtropical Resources and Environment, 3*(3):53–58(In Chinese).
56. Huang X. H., Yu F. K. (2013): Analysis on main nutrients in surface soils from plantation of six *Eucalyptus* species Fumin county. *Modern Agricultural Science and Technology, 8*:191–193 (In Chinese).
57. Zhang X.X., Li Z.B., Li P.(2010): Loess plateau grassland soil trace element distribution characteristics research. *Journal of soil and water conservation, 24*(5):45–48 (In Chinese).
58. Zhong J. H., Li S. Y., Lan P. L., Liao X. R., Liao G. R. (2005): Characteristics and Causes of Soil Fertility Under *Eucalyptus* Plantations in Leizhou Peninsula. *Bulletin of Soil and Water Conservation, 25*(3):44–48 (In Chinese).
59. Yuan Y. H., Fan H. B., Huang R. Z., Su B. Q., Liu W. F. (2009): The spatial distribution and influencing factors of soil trace elements of *Eucalyptus* plantation in continuous age sequences. *Ecology and Environment, 18*(1): 268–273 (In Chinese).
60. Zhao X. Q., Yang S. H. (2008): Research on the Microelements in the soil of the main vegetation types in the subtropical mountainous zone of southwest Yunnan. *Research of Soil and Water Conservation, 5*(15):140–144 (In Chinese).
61. Guo X. D., Fu B. J., Chen L. D., Ma K. M., Li J. R. (2001): Effects of Land Use on Soil Quality in a Hilly Area--A Case Study in Zunhua County of Hebei Province, *Acta Geographica Sinica, 56*(4): 447–455 (In Chinese).
62. William T., David L. (1985): Nutrient dynamics in a agricultural watershed observations on the role ariparian forest. *Ecology, 65*(5): 1466–1475.
63. Qiu L. P., Zhang X. C. (2006): Effects of Land Use on Soil Properties in Ziwuling Region. *Journal of Natural Resouces, 21*(6): 965–972 (In Chinese).
64. Dunjó G., G. Pardini, Gispert M. (2003): Land use change effects on abandoned terraced soils in a Mediterranean catchment, NE Spain. *Catena, 52*( 1):23–37.
65. Gong J., Chen L. D., Fu B. J., Li Y. M., Huang Z. L, Huang Y. L., Peng H. G. (2004): Effects of land use and vegetation restoration on soil quality in a small catchment of the Loess Plateau. *Chinese Journal of Applied Ecology,15*( 12) : 2292–2296 (In Chinese).
66. Xia T. Y., Duan C. Q., Zhang C. X., Fu D. G., Duan Z. Y., Da L. G. (2010): A study on the soil fertility in *Eucalyptus* robusta plantation and their adjacent vegetation. *Journal of Yunnan University, 32*(1):118–123 (In Chinese).
67. Sili P. (2013): Anqiang Chen.Haidong Fang. Effects of vegetation restoration types on soil quality in Yuanmou dry-hot valley, China. *Soil Science and Plant Nutrition, 59*(3):347–360.
68. Wang J. J. (2011): Variation characteristics of soil quality in *Eucalyptus* plantations: [Dissertation]. Nanjing: Nanjing forestry University (In Chinese).
69. Zhong M. Y., Huang S. C., Yang M. S., Chen S. X., Wu Z. H. (2006): Research on soil fertility of *E.urophylla* × *E.grandis, Pinus massoniana* Lamb and *Acacia mangium* plantations. *Eucalypt Science and Technology, 23*(2):33–37 (In Chinese).

# Pollution Status of Selected Metals in Surface Sediments of the Pearl River Estuary and Daya Bay, South China Sea

LINGLONG CAO[1,2], CHUGUANG HUANG[3], JIANHUA WANG[1,*], JIAN XIE[2,*], ZHIXIN NI[3], GANGXIONG JIN[4],
LALI WAXI[1] and HUIXIAN CHEN[1]

[1]*School of Earth Sciences and engineering geology, Guangdong Provincial Key Laboratory of Geological Processes and Mineral Resource Exploration, SunYat-sen University, Guangzhou Guangdong 510275, PR China*
[2]*South China Sea Marine Engineering and Environment Institute, SOA, Guangzhou Guangdong 510300, PR China*
[3]*South China sea marine Environment Monitoring Center, SOA, Guangzhou Guangdong 510915, PR China*
[4]*Taizhou municipal bureau of land and resources Huangyan branch, Taizhou Zhejiang 318200,PR China*

**ABSTRACT:** Selected metals (Cr, Ni, Cu, Pb, Zn, Cd and As) in surface sediments from the Pearl River Estuary (PRE) and Daya Bay (DYB) were analyzed to observe their total concentrations and chemical phase portioning. It was shown that the amounts of all contents, except Pb, have decreased over the past decade. The mean concentrations in PRE, excluding Pb, were significantly higher than those in DYB. Based on the *PLI* and mERM-Q, it was revealed that the highest potential ecological risk zones were in the northwest of PRE and sub-basins of DYB. An overwhelming majority of sediments had a 21% probability of toxicity in PRE and 9% probability of toxicity in DYB. The speciation analyzed by the optimized BCR sequential extraction method showed that, in both areas, Cr, As and Ni were present dominantly in the residual fraction and thus of low bioavailability, while Cd and Pb were found to be abundant in the non-residual fraction and thus of high potential availability, indicating significant anthropogenic sources. The partitioning of Cu and Zn showed a significant difference between the two areas. The source analysis shows that the pollutants resulted primarily from anthropogenic material, and secondly from the input of natural weathering products in PRE, while being mainly attributed to natural geological sources of metals, followed by industrial wastewater and aquaculture activity in DYB.

## INTRODUCTION

As the most important locations for human habitation, coastal and estuarine zones are often polluted by multifarious contaminants, including trace metals. These have drawn particularly great attention due to their toxicity, persistence and biological accumulation once concentrations surpass certain thresholds in sediments, resulting in potential long-term implications on human health and ecosystems (Duan *et al.,* 2014; Ozbas *et al.*, 2007). The toxic effects and biogeochemical pathways of metals can be studied based on the measurements of these forms. Most previous studies have proven that, under the contamination of sediments, using only the total metal concentrations is insufficient as a criterion to assess the environmental effects of contaminated sediments, which critically depends on the specific chemical species and binding states (Yu *et al.*, 2010). Therefore, in addition to measuring their total concentrations, determining the geochemical fractionation of metals in sediments is equally important for assessing their potential toxicity and threat to organisms.

A large number of sequential extraction procedures have been developed based on the Tessier protocols (Tessier *et al.*, 1979; Huang *et al.* 2013). The optimized three-stage sequential extraction procedure, proposed by the European Community Bureau of Reference (BCR), and based on the process known as fractionation in which a sequential series of selective extract is used, is a useful approach to reveal the different affinity to various solid-phase fraction samples, including freshwater sediments, salt water sediments, soil, sewage sludge, and particulate matter. Furthermore, the method has been widely applied and adopted by a large number of researchers to study the speciation

---

*Authors to whom correspondence should be addressed.
E-mail: adswjh@mail.sysu.edu.cn, Tel./fax:+86 20 84112526
E-mail: scsmeei@163.com, Tel./fax:+86 20 84284359

1544-8053/14/04 119-12

and possible associations between metals and sediment components (Huang *et al.* 2013; Qiao *et al.*, 2013).

Metal contamination and environmental degradation have gradually increased in recent years, due to the rapid boom of the local economy and urban development, thus the Pearl River Delta (PRD), located along the south coast of China, has become one of the most developed regions in the country (Yu *et al.*, 2010). The Pearl River Estuary (PRE) and Daya Bay (DYB) are the most important economic development districts and are among the areas most greatly affected by human activities throughout the PRD, and even all of China. Trace metals in the surface sediments of both areas have ubiquitously accreted over the last two decades, and have revealed a potential contamination problem. To the best of our knowledge, previous studies regarding the surface sediments of both areas have moved their focus from the analysis of total concentrations, to deposition, migration and accumulation mechanism, further toxicity and evaluation. However, investigations related to metal speciation and comprehensive multi-factor characteristics analyses have been scarce. Therefore, the outline of the present study is as follows: (1) to reveal the accumulation, spatial distribution and speciation of the selected metals; (2) to assess the enrichment status and potential ecological risk using several contamination indices; and (3) to use correlation analysis (CA) and principal component analysis (PCA) to define the potential sources of the metals. Further aims are as follows: first, to determine and compare the chemical speciation of the metals to evaluate their relative mobility and bioavailability, then to determine, where possible, the contributing origin of the selected metals, with the aid of multivariate statistical analyses.

## MATERIAL AND METHODS

### Sampling and Sediment Pretreatment

32 and 22 surface sediment samples (0–5 cm) were collected in triplicate, respectively from PRD and DYB, in May 2011, using a stainless steel grab sampler and a small vessel, then sealed in polyethylene bags and stored at low temperature (–20°C). A map of the sampling locations is shown in Figure 1. Upon analysis, samples were defrosted and air-dried in an oven at 60°C to constant weight, then ground to a powder using an agate mortar and pestle, and sieved to collect the < 63 μm grain size fraction for homogenization, which has a strong association with metals (Tam and Wong, 2000). Approximately 0.5 g of dried and homogenized

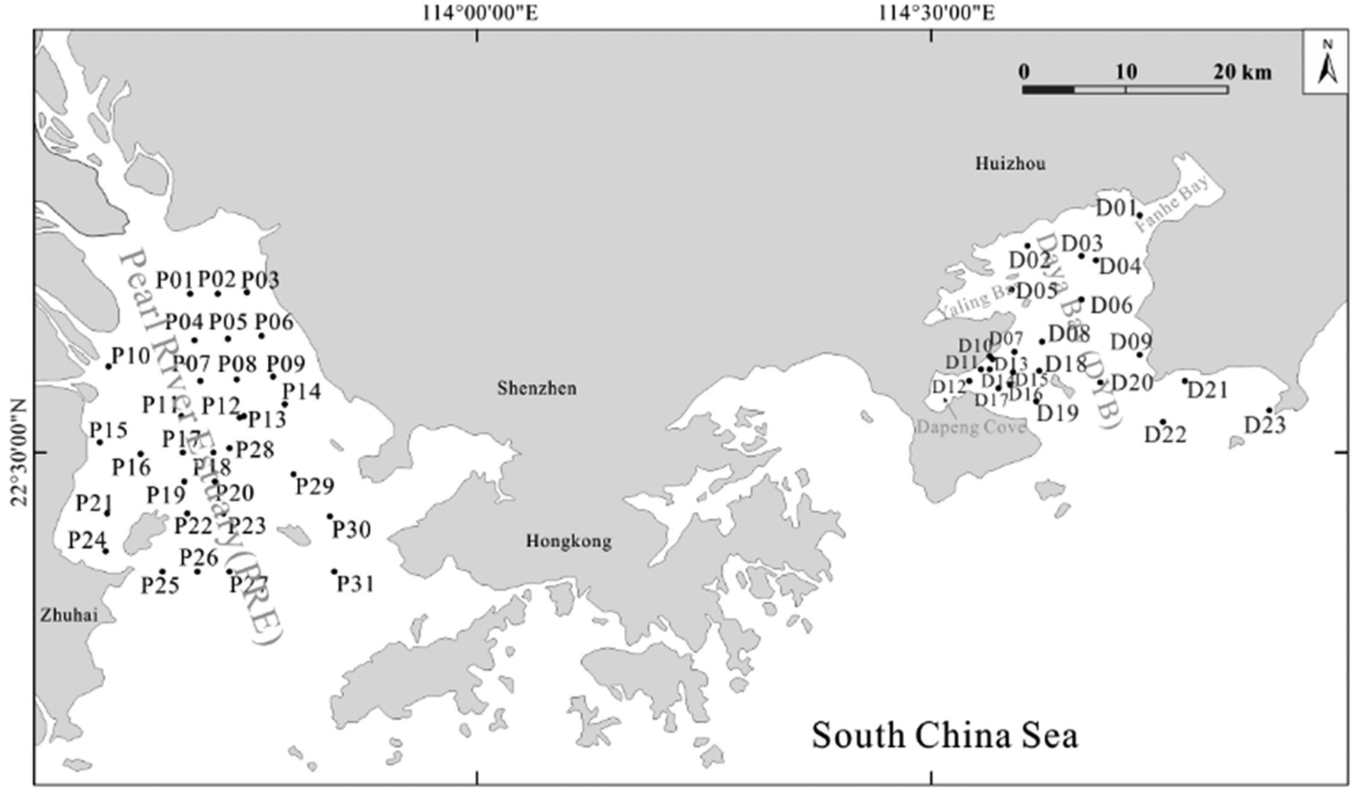


***Figure 1.*** *Locations of sampling stations of the surface sediments from the studied regions.*

sediment samples were weighed and placed into an acid washed PTFE digestion, using the established US EPA method 200.2 protocols (USEPA, 1994).

### Total Concentrations and Phase Partitioning of Metals

The concentrations of Cr, Ni, Cu, Pb, Zn, Cd and As were all detected by means of inductively coupled plasma-mass spectrometry (ICP-MAS). Quality control was assured through the analysis of a marine sediment reference material (Offshore Marine Sediment, GBW 07314) and duplicate samples. The analysis results were deemed reliable when repeat sample analysis error was below 5%, and the analytical precision for replicate samples was within ±10%. The optimized BCR sequential extraction procedure reported by Rauret *et al.* (1999) was used to partition the metals into four geochemical parts as exchangeable (F1), reducible (F2), oxidizable (F3) and residual (R) fractions. The detailed procedures for the sequential extraction used in this study have been described elsewhere (Huang *et al.*, 2013). The recovery rates for all of the metals were 85–115%.

### Statistical Analysis

Pearson correlation and multivariate Principal Component Analysis (PCA) were performed on IBM SPSS 19.0 for Windows. When $p < 0.05$ it was considered that a significant difference was present. PCA was performed on the standardized dataset to minimize the effects of the differences in the measurement units or variance and to render the data dimensionless, then to analyze and identify the possible sources of metal contamination in the studied area and evaluate the degree of association among the variables.

## RESULTS AND DISCUSSION

### Metal in Total Concentrations

The large spatial distribution variation of metals in the sediments of the PRE is presented in Figure 2, which reveals a trend of gradually decreasing from the northwest to the southeast. The results show that the metal concentrations in the sediments from the west side of PRE were higher than those from the east side. This is in accord with previous reports (Ye *et al.*, 2012), which attributed the higher metal concentrations in the west shallow water areas to the direct river input from the north-western side, and the hydraulic conditions which may facilitate sedimentation. According to Figure 2, the distribution features of metal concentrations in DYB show a ring shape, and descend from the shore to bay by varying degrees. Relatively higher concentrations of the selected metals, except Cr, occurred in the nearby sub-basins of DYB, where frequent human activities were present, including high value areas for Cu, Pb, Zn, Ni and Cd near Dapeng Cove and Yaling Bay, and the maximum zone for As appeared in Fanhe Bay. The reason for this distribution pattern may be associated with emissions from the human activities in or near these sub-basins, which may make significant contributions to the enrichment of these metals and are relatively consistent with a year-round clockwise circulation in DYB. In general, the mean values of all of the selected metals, except Pb, were clearly higher in the sediments of PRE than those of DYB, especially for Cd, Cu and As. The mean values for Cd (0.66 mg kg$^{-1}$), Cu (43.07 mg kg$^{-1}$) and As (14.09 mg kg$^{-1}$) in the sediments of PRE were respectively 18 times, four times and twice as high as those in DYB, which is consistent with the results of previous studies (Yu *et al.*, 2010). Only for Pb, the average content (41.38 mg kg$^{-1}$) in PRE was slightly lower than in the sediments of DYB (44.18 mg kg$^{-1}$). Clearly, different from the well preserved semi-closed DYB which does not have land runoff input, the PRE receives large quantities of terrestrial materials, especially from the mainstream of the Pearl River, thus leading to the relatively higher inputs of anthropogenic metals there (Duan *et al.*, 2014).

A comparison of data set revealed that the mean concentration of the metals, except Pb, showed decrease year by year in the studied areas (Table 1). In the surface sediments from the PRE, the mean values of Cr, Ni, Pb and Zn respectively decreased by 2.6, 1.6, 1.4, and 1.4 times in the past decade, and the highest value of Cr significantly dropped from 89 mg kg$^{-1}$ to as low as 33.86 mg kg$^{-1}$. In DYB, our most widespread elements, including Cr (30.03 mg kg$^{-1}$), Ni (18.95 mg kg$^{-1}$), Cu (10.9 mg kg$^{-1}$), Zn (59.34 mg kg$^{-1}$) and As (7.01 mg kg$^{-1}$), also have relatively lower concentrations than those found in former studies, e.g. Yu *et al.* (2010) and Qiu *et al.* (1997), but only Pb (44.18 mg kg$^{-1}$) is noticeably higher than that of the past decade (23.00 mg kg$^{-1}$). Among all of the coastal bays chosen for comparison (Table 1), the average contents of Cu, Pb, Cd in PRE and Pb in DYB were higher than those of the Yangtze River Estuary and Bohai Bay

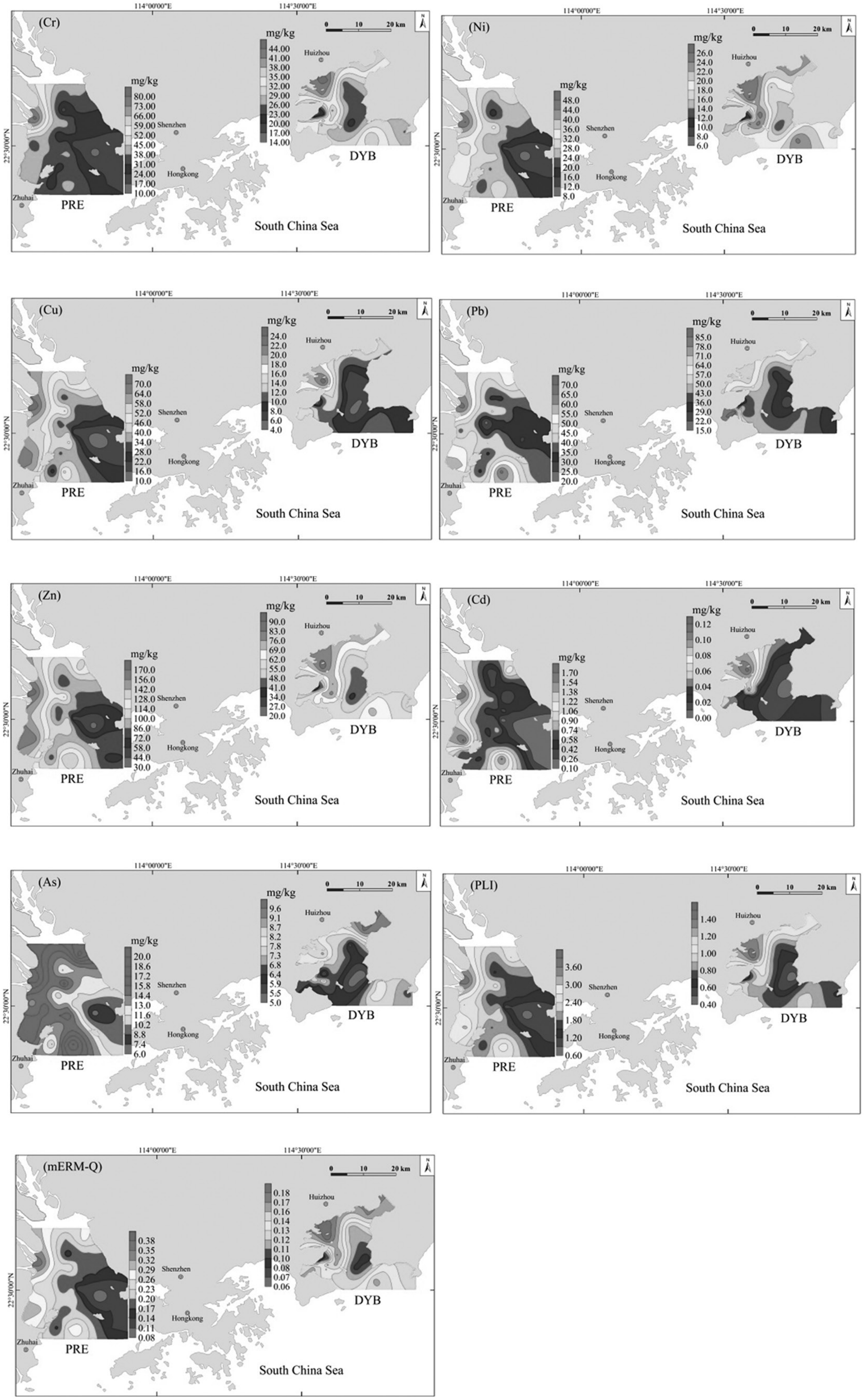


***Figure 2.*** *Spatial distribution of selected metals concentrations PLI and mERM-Qin surface sediments from the studied regions.*

***Table 1. Metal Concentration in Surface Sediments from the Studied Regions Compared with the Average Metal Concentration in Sediments of Other Bays (mg/kg, dry wt).***

| Location | | Cr | Ni | Cu | Pb | Zn | Cd | As | References | Sample Date |
|---|---|---|---|---|---|---|---|---|---|---|
| Pearl River Estuary | range | 14.67~82.30 | 9.90~48.29 | 10.66~173.74 | 22.69~66.99 | 37.79~275.11 | 0.11~1.75 | 6.79~20.47 | this study | 2011 |
| | average | 33.86 | 25.85 | 43.07 | 41.38 | 104.65 | 0.66 | 14.09 | | |
| | | NA | NA | NA | 40.51 | 109.09 | 0.29 | 17.42 | Ye *et al.* (2012) | 2009 |
| | | 107.00 | 37.10 | 44.00 | 54.70 | 177.00 | NA | NA | Zhang *et al.* (2009) | 2008 |
| | | 89.00 | 41.70 | 46.20 | 59.30 | 150.00 | NA | NA | Zhou *et al.* (2004) | 1999 |
| Daya Bay | range | 14.82~44.69 | 6.62~26.15 | 4.27~23.51 | 18.68~89.58 | 22.34~87.25 | 0.002~0.114 | 5.15~9.73 | this study | 2011 |
| | average | 30.03 | 18.95 | 10.90 | 44.18 | 59.34 | 0.04 | 7.01 | | |
| | | 75.40 | 25.60 | 10.50 | 32.70 | 96.70 | NA | NA | Yu *et al.* (2010) | 2008 |
| | | NA | NA | 25.00 | 23.00 | 89.00 | 0.03 | 9.60 | Qiu *et al.* (1997) | 1996 |
| Yangtze River Estuary | average | 78.90 | 31.80 | 30.70 | 27.30 | 94.30 | 0.26 | NA | Zhang *et al.* (2009) | 2005 |
| Quanzhou bay | | 82.00 | NA | 71.40 | 67.70 | 179.60 | 0.69 | 21.70 | Yu *et al.* (2008) | 2006 |
| Bohai bay | | 101.00 | 40.70 | 38.50 | 34.70 | 131.00 | 0.22 | NA | Gao *et al.* (2012) | 2008 |
| Masan Bay,Korea | | 67.10 | 28.80 | 43.40 | 44.00 | 206.30 | 1.24 | NA | Hyun *et al.* (2007) | 2005 |
| Aliağa bay, Turkey | | 111.00 | 98.70 | 321.00 | 284.00 | 86.40 | 1.47 | NA | Neser *et al.* (2012) | 2010 |
| BMEC[a] | | 39.30 | 22.10 | 7.43 | 15.60 | 54.40 | 0.18 | 9.71 | Zhang *et al.* (2005) | 1998 |
| SQG of USEPA | ERL[b] | 81.00 | 20.90 | 34.00 | 47.00 | 150.00 | 1.20 | 8.20 | Long *et al.*,1995 | |
| | ERM[c] | 370.00 | 51.60 | 270.00 | 218.00 | 410.00 | 9.60 | 70.00 | | |

[a] BMEC: Background of marine sediments in continental shelf of South China Sea.

[b] ERL, Effects Range-Low, threshold below which adverse effects on biota are rarely observed.

[c] ERM, Effects Range-Median, threshold above which adverse effects on biota are frequently observed.

(Zhang *et al.*, 2009), but much lower than those of Quanzhou Bay (Yu *et al.*, 2008). For the other three selected metals (Cr, Ni, As), the mean contents in this studied areas were lower than those in all the other domestic coastal bays listed in Table 1, while the average value of Zn (104.65 mg kg$^{-1}$) in PRE, which was also lower than the contents in Quanzhou Bay and Bohai Bay, was 10% higher than that in the Yangtze River Estuary. In addition, all selected metals in the studied areas were significantly lower than those in other larger industrialized urban bays throughout the world, e.g. Masan Bay of South Korea (Hyun *et al.*, 2007) and Aliafia Bay of Turkey (Neser *et al.*, 2012). However, compared with the corresponding background values in the South China Sea, the surface sediments collected were dramatically contaminated by the selected metals, except Cr, in the PRE and Cu, Pb and Zn in DYB.

## Metal Enrichment and Potential Risk

The pollution load index (*PLI*) was introduced to evaluate the level of metal contamination. The *PLI* is determined as the *n*th root of the *n* contamination factors ($CF_n$) multiplied together and calculated using the following equation:

$$PLI = (CF_1 x\ CF_2 x\ CF_4 x \ldots\ldots CF_n)^{1/n} \qquad (1)$$

The *CF* for each metal is the ratio of the measured concentration in the sediment to the background value, which is classified into four grades for monitoring the pollution of one single metal over a certain period of time.

The *PLI* was applied to determine the integrated pollution status of the combined toxicant groups at the sampling stations. The *PLI* value > 1 indicates a polluted condition, while *PLI* < 1 signifies no metal pollution (Ozbas *et al.,* 2007). The spatial map of *PLI* in Figure 2 shows that the contamination distribution pattern was in accord with the concentration distribution as a whole, and the contaminated risk was higher in PRE than DYB. In PRE, except for station P29 which had a *PLI* value of 0.75, the *PLI* results for all the other stations ranged from 1.07 to 3.81, and the highest *PLI* value of 3.81 occurred at station P10, making this the most contaminated site, followed in order by P21, P27, P3, P26 and P24, the respective values of which were 3.04, 2.85, 2.71, 2.58 and 2.62. In comparison with PRE, the *PLI* values were clearly lower in DYB, less than 1.0 for all stations except for D3 (1.48), D4 (1.30), D10 (1.07), D15 (1.05), D21 (1.05), suggesting that contamination areas were present in the vicinity of sub-basins.

Effects Range-Low (ERL) and Effects Range-Median (ERM) were the main parameters used to estimate the adverse biological effects of metals in the marine and estuary sediments and derived for the US National Oceanic and Atmospheric Administration (NOAA), with numerous types of modeling, laboratory and field studies performed on marine and estuarine sediments (Long *et al.*, 1995). Metal concentrations in sediments can be classified into three levels, namely rarely (< ERL), occasionally (ERL-ERM) and frequently (≥ ERM) associated with adverse biological effects. Our results (Table 1, Figure 2) show that ERM was never exceeded in the study areas, and even in DYB never exceeded ERL, which would likely not cause adverse effects in the studied areas. The Ni, Cu and As were between the ERL and ERM values in PRE, indicating that adverse effects on benthic organisms are occasionally observed. The results suggest that Ni, Cu and As in the northwest of PRE could be given more attention, due to the potential environmental risks they may cause.

All of the indexes applied above took into account the individual metals. Based on the fact that metals always occur in sediments as complex mixtures, the mean ERM quotient method was used to determine the possible biological effects of the combined toxicant groups, by computing the mean quotients for a large range of contaminants (Carr *et al.*, 1996). The mean ERM quotient is calculated using the following formula:

$$\text{mean-ERM quotient} = \Sigma(C_x/\text{ERM}_x)/n \qquad (2)$$

where $C_x$ is the measured concentration of the examined component ($x$) in the sediment, ERM*x* is the ERM for metal *x*, and *n* is the number of metals. The mean-ERM quotients of less than or equal to 0.1 (low priority site) have a 9% probability of being toxic, quotients of 0.11–0.5 (medium-low priority site) have a 21% probability, quotients of 0.51–1.5 (high-medium priority site) have a 49% probability, and quotients of greater than 1.50 have a 76% probability (Long *et al.*, 2000). Based on the mERM-Q data, a contour map of multi-metals ecological risk potential is presented (Figure 2). In the surface sediments of the studied regions, all the investigated stations had mERM-Q < 0.5 varying within the range of 0.06–0.37. In general, all sites except P29 in PRE were medium-low priority sites, indicating that the combination of the seven studied metals have a

21% probability of being toxic. In contrast, the majority of the investigated stations in DYB had mERM-Q < 0.1 (low priority site), and had a 9% probability of being toxic. Both the *PLI* and mERM-Q revealed that the highest potential ecological risk zone was found in the northwest of PRE.

## Metal Speciation and Correlation Analysis

It has been demonstrated that metal speciation and solubility affect the mobility, bioavailability and toxicity of metals in sediments which largely depend on the physiochemical form (Tessier *et al.*, 1979; Yu *et al.*, 2010). Therefore, the optimized BCR sequential extraction technique was applied to the sediments collected from the research areas, and the percentages of metals in the sediment samples from each extraction step are presented in Figure 3. The selected metals take on different speciation patterns, and some show significant spatial variation.

The results in Figure 3 show that the chemical partitioning of the selected metals (Cr, Ni, Cu, Pb, Zn, Cd, As) in the sediment were variable in both areas. Cr, As and Ni of both areas were dominant in the residual fraction, which can be considered as the most environmentally safe component, while Cd and Pb were mostly abundant in the acid-soluble fraction and reducible fraction, respectively, indicating that they are either more or less available to aquatic biota and may correlate with their concentrations in microorganisms. The fractionation patterns of Cu and Zn showed significant differences in both regions, which were mostly presented in reducible form in PRE and acid-soluble form in DYB.

It is widely known that the non-residual proportion reflects the reactivity of metals to some extent, namely the higher the proportion of heavy metal is, the stronger the activation is, and the easier the remobilization becomes. Based on the concentrations of fractions in each step of extraction, the sequence in the proportion of non-residual form is in decreasing order, as follows:

PRE: Cd>75%>Pb>Cu>Zn>Ni>50%>Cr>As>20%

DYB: Pb>Cd>75%>50%>Cu>Zn>Ni>Cr>As>25%

It has been well established that Cd is a typical anthropogenic metal and mostly enters the aquatic environment through the discharge of industrial and domestic effluents which are more easily remobilized than metals naturally present in the environment. The results of the present study show that Cd in the non-residual proportion was greater than 75%, even as high as 85% in PRE, which indicates that Cd has strong activity in natural environments, and the pollution status of Cd in the study areas is becoming more and more serious due to anthropogenic influence. From Figure 3, the results in PRE show that Cd in the acid-soluble fraction is the most dominant form, accounting for an average of 75.99% of its mean total concentration, reaching a maximum of 82.71%, and its percentages in the oxidizable fractions were almost negligible. Previous research has also shown that Cd was particularly prone to remobilization from sediment towards the greater bioavailability in the Mai Po mangroves, river mouths of Hong Kong (Man *et al.,* 2004), and southern area of PRE, and presented a high risk to the environment with a high percentage in the acid-soluble fraction. The variation of Cd speciation was the most significant in DYB, indicating its possible point source input feature, as 78.65% of the mean total concentration of Cd was found in the non-residual fraction, while only 4% of total Cd was found in the fractions at D8. The region contains major aquiculture areas and busy ports, thus the use of phosphate fertilizer and corrosion resistant painting from the boats frequently transporting cargo in the bay and berthing in the dock may be among the critical sources of the local Cd pollution.

It is potentially bioavailable that the prevailing phase of Pb was in the non-residual fraction accounting for 78.83% in DYB, and the dominant phase of Pb was in the reducible fraction in PRE, which accounts for 46.04–70.12% of its total content, with an average of 60.24%. Moreover, the percentages of it in acid-soluble and oxidizable fractions are almost equal, with respective averages of 6.29 and 6.56%. Lead is highly toxic to the environment. The adsorption of Pb cations on the hydrous (amorphic) oxides of Fe/Mn is considered as a universal fixation mechanism, and the ability of lead to form organic complexes is widely known (Qiao *et al.*, 2013). In the areas influenced by extensive human activities, a considerable proportion of Pb in the environment is determined to be of anthropogenic origin, as it is an important additive which has been widely used in many types of products. The reducible Pb predominating in non-residual fraction has been reported by many researchers (Ip *et al.*, 2007). The speciation of Pb shows no significant spatial variation in the surface sediments of PRE, indicating a non-point source input feature.

Cu speciation was found to be highly variable and the non-residual fraction of Cu in PRE accounts for

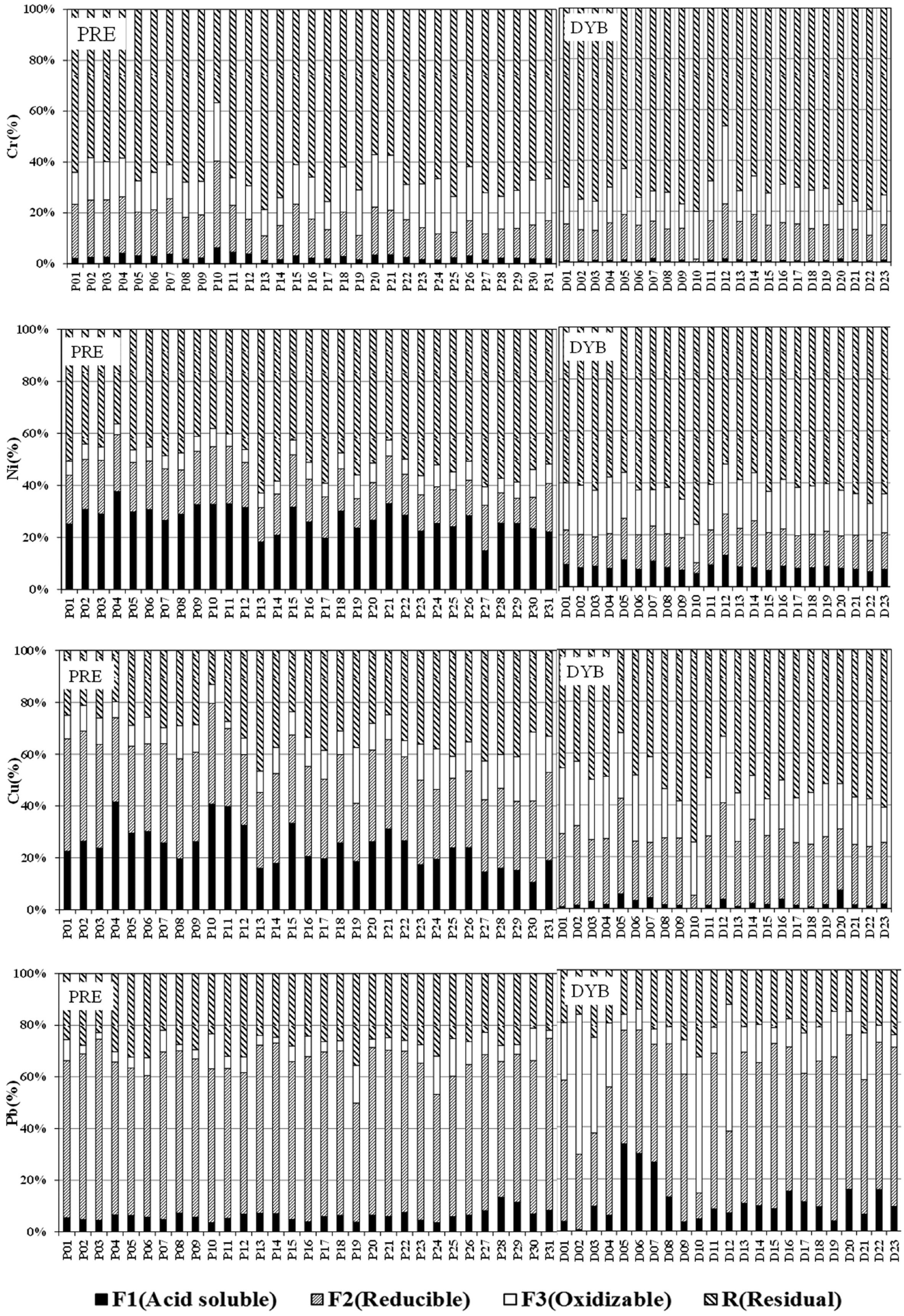


***Figure 3.*** *Distribution in different geochemical fractions of surface sediments from PRE (left) and DYB (right).*

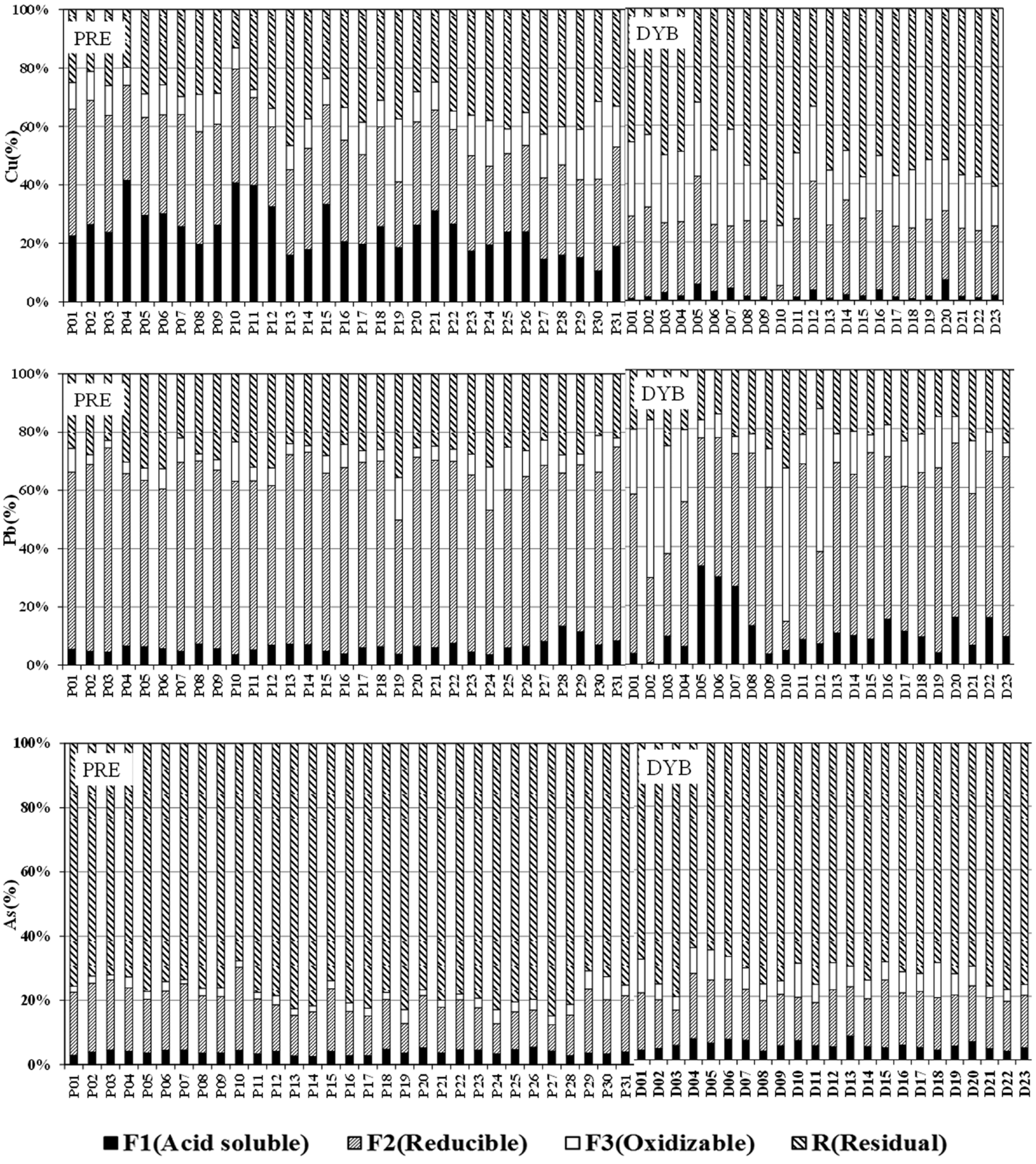


***Figure 3 (continued).*** *Distribution in different geochemical fractions of surface sediments from PRE (left) and DYB (right).*

54–89% of its total content, with an average of 69.31%. The proportion to non-residual fraction was almost equal in DYB, the mean value of which is 49.54%. In addition, the partitioning of Cu showed significant differences in the research areas. The percentage of Cu in the non-residual fractions was found to be clearly higher in the sediments in PRE than in DYB, which may be due to the influx in industry sewage and domestic wastewater in PRE. Similar variation patterns also occurred in the Zn speciation in the DYB sediments of the sampling regions. These observations indicate that Cu and Zn in the sediments of the PRE were more mobile and freshly deposited and when the pH value and reducing condition changed in the sediments, as this may more easily result in secondary pollution.

The characteristics that both areas had in common were that the major quantify of As, Cr and Ni were found to be associated with the residual fraction, the mean values of which were 77.31, 65.55 and 51.01% in PRE, and 72.24, 71.57 and 61.69% in DYB. The relative content of a metal in the residual fraction can be used as a measure of the contribution of natural sources, and a higher percentage in this fraction indicates a lower level of pollution and the mineralogical origin of the metal in sediments of the sampling regions. Therefore, the high percentage of As, Cr and Ni in the residual fraction indicated that they were relatively stabilized and had a low potential bioavailability and toxicity.

The risk assessment integrating the geochemical fractionation and total content of metals is vital for the understanding of the pollution level of heavy metals for the better management practices of coastal sediments (Yu *et al.*, 2008). The correlation matrix between the geochemical phases and total content of metals in the study areas are shown in Table 2. The results also show that, except Pb (F1) and As (F3) in PRE and Cr (F2), Pb (F1), Cd (R), As (F1) in DYB, the concentrations of all metals in both areas were positively and highly correlated with their metal speciation, indicating that it was reliable to some extent to determine the level of pollution by the total concentration of heavy metal.

## Principal Component Analysis (PCA)

Previous studies regarding chemical fractionation were usually carried out using univariate procedures; however, multivariate methods such as Pearson correlation and multivariate principal component analysis (PCA) may provide further statistical interpretation of the results. To explore the correlations among the concerned variables, the Pearson correlation analysis was performed and the results are shown in Table 2. Significantly positive correlations were observed for selected metals, except for Cd and As in DYB, indicating that these metals are associated with each other and may have common natural or anthropogenic sources (Tam, 2010). PCA was performed on the metal fractionation data, and the scores in the rotated principal component space are presented in Table 3. The results obtained from PCA were confirmed by the correlation analysis and were applied to the data sets (28 variables) separately for the two typical study areas to identify the sources of metals in the sediments, by employing varimax rotation with Kaiser Criterion.

PCA in the PRE data set involves three PCs with eigenvalues > 1, explaining approximately 77.9% of the information contained in the total variables, which is sufficient to provide information for the data structure. The first PC (PC1) accounts for 31.79% of the total variance and presents high loading values on acid-soluble fraction (Cr, Ni, Cu, Zn, Cd, As) and reducible fraction (Cr, Ni, Cu, Zn, Cd, As). Therefore, based on the weight of the contribution of each variable, it can be found that both F1 and F2 (positive loading) were extremely highly correlated with PC1, and the results also suggest that PC1 has different geochemical behaviors and is mainly sourced from mixed anthropogenic inputs, including industrial waste (such as metallurgy, manufacturing, chemical industry) and sewage, which is in accord with the above-mentioned geochemical fractions analysis. The second PC (PC2), which accounted for 30.34% of the total variance with high positive loading on the residual forms (Cr, Ni, Cu, Zn, Cd, As), infer that it mainly originates from mineral weathering. PC3 explains 15.78% of the total variance and presents good loading values for the oxidizability of Ni, Cu, Zn, Pb and As; most of these ingredients were combined with organic matter, thus it can be inferred as a biological activity factor.

In DYB, two components were extracted which explain 71.10% of the total variance, and the eigenvalues > 1. PC1 accounted for 48.58% of the total variance and was characterized by high loadings of residual fraction for almost all selected metals and reducible fraction from Cr, Ni, Zn, Pb and As, revealing that they reflected the contribution of the natural geological sources of

***Table 2. Correlation Matrix Between Total Concentration and Geochemical Phases of Selected Metals in Studied Areas.***

| | Variables | Cr | Ni | Cu | Pb | Zn | Cd | As |
|---|---|---|---|---|---|---|---|---|
| PRE | F1 | 0.866** | 0.879** | 0.963** | 0.361 | 0.956** | 0.998** | 0.851** |
| | F2 | 0.894** | 0.896** | 0.982** | 0.937** | 0.946** | 0.979** | 0.644** |
| | F3 | 0.946** | 0.836** | 0.779** | 0.687** | 0.756** | 0.937** | 0.197 |
| | R | 0.813** | 0.857** | 0.710** | 0.828** | 0.780** | 0.662** | 0.967** |
| DYB | F1 | 0.334 | 0.815** | 0.693** | 0.345 | 0.418* | 0.777** | 0.131 |
| | F2 | 0.810** | 0.927** | 0.964** | 0.657** | 0.957** | 0.686** | 0.771** |
| | F3 | 0.678** | 0.940** | 0.910** | 0.718** | 0.710** | 0.664** | 0.566** |
| | R | 0.915** | 0.975** | 0.818** | 0.775** | 0.941** | 0.184 | 0.933** |

*Correlation is significant at the 0.05 level (2-tailed).

**Correlation is significant at *P* the 0.01 level (2-tailed).

***Table 3. Pearson Correlation cCoefficients Between Heavy Metals in Studied Regions.***

| PRE | | | | | | | |
|---|---|---|---|---|---|---|---|
| | **Cr** | **Ni** | **Cu** | **Pb** | **Zn** | **Cd** | **As** |
| **Cr** | 1 | | | | | | |
| **Ni** | 0.941** | 1 | | | | | |
| **Cu** | 0.964** | 0.869** | 1 | | | | |
| **Pb** | 0.775** | 0.739** | 0.702* | 1 | | | |
| **Zn** | 0.953** | 0.920** | 0.944** | 0.809** | 1 | | |
| **Cd** | 0.721** | 0.774** | 0.674* | 0.725** | 0.832** | 1 | |
| **As** | 0.785** | 0.840** | 0.661* | 0.862** | 0.764** | 0.748** | 1 |

| DYB | | | | | | | |
|---|---|---|---|---|---|---|---|
| | **Cr** | **Ni** | **Cu** | **Pb** | **Zn** | **Cd** | **As** |
| **Cr** | 1 | | | | | | |
| **Ni** | 0.940** | 1 | | | | | |
| **Cu** | 0.830** | 0.726** | 1 | | | | |
| **Pb** | 0.662* | 0.468 | 0.751** | 1 | | | |
| **Zn** | 0.935** | 0.958** | 0.840** | 0.592* | 1 | | |
| **Cd** | 0.382 | 0.257 | 0.722** | 0.559* | 0.450 | 1 | |
| **As** | 0.566* | 0.544* | 0.528* | 0.389 | 0.551* | 0.218 | 1 |

*Coefficients at 0.05 significance level, $p < 0.05$.

**Coefficients at 0.01 significance level, $p < 0.01$.

***Table 4. Loadings in Fractions of Metals on VARIMAX-rotated Ractors of Different Datasets.***

| | PRE | | | DYB | |
|---|---|---|---|---|---|
| **Metals** | **PC1** | **PC2** | **PC3** | **PC1** | **PC2** |
| CrF1 | **0.93** | 0.09 | 0.14 | –0.02 | **0.64** |
| NiF1 | **0.82** | 0.32 | 0.11 | **0.69** | 0.49 |
| CuF1 | **0.94** | 0.09 | 0.19 | 0.03 | **0.90** |
| PbF1 | –0.19 | –0.10 | 0.08 | 0.06 | **0.90** |
| ZnF1 | **0.86** | 0.26 | 0.20 | 0.08 | **0.82** |
| CdF1 | 0.51 | 0.47 | 0.08 | 0.36 | **0.72** |
| AsF1 | 0.45 | **0.60** | 0.06 | –0.43 | 0.31 |
| CrF2 | **0.96** | 0.18 | 0.09 | **0.57** | 0.43 |
| NiF2 | **0.84** | 0.42 | –0.02 | **0.85** | 0.33 |
| CuF2 | **0.93** | 0.27 | 0.15 | 0.47 | **0.59** |
| PbF2 | 0.49 | **0.62** | 0.15 | 0.34 | **0.61** |
| ZnF2 | **0.85** | 0.27 | 0.31 | **0.83** | 0.42 |
| CdF2 | 0.57 | 0.47 | 0.12 | –0.07 | 0.42 |
| AsF2 | **0.87** | 0.29 | –0.23 | **0.62** | 0.21 |
| CrF3 | **0.76** | 0.36 | 0.46 | 0.36 | 0.28 |
| NiF3 | 0.44 | 0.55 | **0.68** | **0.87** | 0.17 |
| CuF3 | 0.49 | 0.46 | **0.70** | 0.30 | **0.61** |
| PbF3 | 0.31 | 0.28 | **0.81** | –0.10 | –0.15 |
| ZnF3 | 0.23 | 0.56 | **0.67** | 0.47 | 0.14 |
| CdF3 | 0.43 | 0.44 | 0.25 | –0.35 | 0.19 |
| AsF3 | –0.14 | 0.10 | **0.91** | 0.12 | 0.13 |
| CrR | 0.3 | **0.87** | 0.28 | **0.91** | –0.01 |
| NiR | 0.25 | **0.89** | 0.32 | **0.98** | 0.04 |
| CuR | 0.32 | **0.78** | 0.43 | **0.86** | 0.13 |
| PbR | 0.29 | 0.55 | 0.59 | **0.52** | 0.07 |
| ZnR | 0.32 | **0.83** | 0.44 | **0.97** | 0.04 |
| CdR | 0.13 | **0.73** | 0.16 | **0.56** | 0.01 |
| AsR | 0.23 | **0.89** | 0.17 | **0.59** | –0.25 |
| Percentage of total variance | 31.79 | 30.34 | 15.78 | 48.58 | 22.52 |
| Cumulative percentage variance | 31.79 | 62.13 | 77.91 | 48.58 | 71.10 |

metals (e.g. weathered bedrock and corrosion product) into the coastal sediments. PC2 accounted for 22.52% of the total variance and was strongly associated with F1 (Cr, Cu, Zn, Cd, Pb), F2 (Cu, Pb) and F3 (Cu).The research results show that industrial wastewater often contains large amounts of a variety of phase metal elements. The DYB includes a large area of farming land, and the contribution toward the high proportion of Pb in F1 and F2 may be mainly related to cage culture activities (feeding fodder) and fishing boats. Therefore, PC2 may be deemed as the industrial wastewater and aquaculture activity factor.

## CONCLUSION

The purpose of this study was to assess the pollution status of some selected metals (Cr, Ni, Cu, Pb, Zn, Cd and As)using a combination of multivariate statistical analyses, sequential extraction data and the ecological risk indexes in surface sediments collected from the Pearl River Estuary and Daya Bay.

According to the results based on the *PLI* and mERM-Q, it was revealed that the priority metal pollutants of concern were in the northwest of PRE and sub-basins of DYB. The above observation was also supported by effects-based SQGs. The mERM-Q demonstrated that the combination of the seven selected metals in sediments of overwhelming majority of the stations had a 21% probability of toxicity in PRE and 9% probability of toxicity in DYB. They were found Cr, As and Ni mostly in the residual fraction with low mobility and thus of low bioavailability to organisms, while Cd and Pb in the non-residual fractions with high bioavailability and contamination risk. The PCA were conducted and show contamination source primarily from anthropogenic material (including industrial wastewater, sewage, etc.), and secondly from the input of natural weathering products in PRE, while being mainly attributed to natural geological sources of metals (e.g. weathered bedrock and corrosion products), followed by industrial wastewater and aquaculture activity in DYB.

The study suggested that for better understanding and preventing heavy metal pollution in an area, continuous environmental monitoring and further geochemical studies should be performed, and supporting the development strategies of contamination control and management.

## ACKNOWLEDGEMENTS

This work was supported by the Ocean Public Welfare Scientific Research Project, State Oceanic Administration People's Republic of China (Grant No. 201105015-06; No. 201105024).

## REFERENCES

Carr, R.S., Long, E.R., Windom, H.L., Chapman, D.C., Sediment quality assessment studies of Tampa Bay, Florida, *Environ. Toxicol. Chem.,* Vol. 15, 1996, pp. 1333–1346.

Duan, D.D., Ran,Y., Cheng, H.F., Chen, J.A., Contamination trends of trace metals and coupling with algal productivity in sediment cores in Pearl River Delta, South China, Chemosphere, Vol.103, 2014, pp. 35–43.

Filgueiras, A.V., Lavilla, I., Bendicho, C., Chemical sequential extraction for metal partitioning in environmental solid samples, *J. Environ. Monit.,* Vol. 4, 2002, pp. 832–857.

Hakanson, L., An ecological risk index for aquatic pollution control. a sedimentological approach, *Water Research,* Vol. 14, 1980, pp. 975–1001.

Huang,L.L.,Pu,X.M.,Pan,J.F.,Wang,B., Heavy metal pollution status in surface sediments of Swan Lake lagoon and Rongcheng Bay in the northern Yellow Sea, *Chemosphere,* Vol. 93, 2013, pp. 1957–1964.

Hyun, S., Lee, C.H., Lee, T., Choi, J.W., Anthropogenic contributions to heavy metal distributions in the surface sediments of Masan Bay, Korea, *Mar. Pollut. Bull,* Vol. 54, 2007, pp. 1059–1068.

Ip, C.M., Li, X.D., Zhang, G., Wai, O.W.H., Trace metal distribution in sediments of the Pearl River Estuary and the surrounding coastal area, South China, *Environ. Pollut.,* Vol. 147, 2007, pp. 311–323.

Long, E.R., Macdonald, D.D., Smith, S.L., Calder, F.D., Incidence of adverse biological effects within ranges of chemical concentrations in marine and estuarine sediments, *Environ. Manage.,* Vol. 19, 1995, pp.81–97.

Long, E.R., MacDonald, D.D., Severn, C.G., Hong, C.B., Classifying the probabilities of acute toxicity in marine sediments with empirically-derived sediment quality guidelines, *Environ. Toxicol. Chem.,* Vol. 19, 2000, pp. 2598–2601.

Man, K.W., Zheng, J.S., Leung, A.P.K., Lam, P.K.S., Lam, M.H.W., Yen, Y.F., Distribution and behavior of trace metals in the sediment and porewater of a tropical coastal wetland, *Sci. Total Environ.,* Vol. 327, 2004, pp.295–314.

Neser, G., Kontas, A., Ünsalan, D., Uluturhan, E., Altay, O., Darilmaz, E., Küçüksezgin,F., Tekogul, N., Yercan, F., Heavy metals contamination levels at the Coast of Aliaga (Turkey) ship recycling zone, *Mar. Pollut. Bull.*, Vol. 64, 2012, pp. 882–887.

Ozbas, E.E., Ozcan, H.K., Balkaya, N.,Demir, G., Bayat, C., Heavy metal amounts in soil and sediments of surface water sources in the industrial regions of Istanbul, *J. Resid. Sci. and Tech.*, Vol. 4, No. 4, 2007, pp.89–94.

Qiao, Y.M., Yang, Y., Gu, J.G, Zhao, J.G., Distribution and geochemical speciation of heavy metals in sediments from coastal area suffered rapid urbanization, a case study of Shantou Bay, China, *Mar. Pollut. Bull.,* Vol. 68, 2013, pp. 140–146.

Rauret, G., Lopez-Sanchez, J.F., Sahuquillo, A., Rubio, R., Davidson, C., Ure, A., Quevauviller, P., Improvement of the BCR three step sequential extraction procedure prior to the certification of new sediment and soil reference materials, *J. Environ. Monit.,* Vol. 1, 2013, pp.57–61.

Tam, N.F.Y., Wong, Y.S., Spatial variation of heavy metals in surface sediments of Hong Kong mangrove swamps, *Environ. Pollut.,* Vol.110, 2010, pp.195–205.

Tessier, A., Campbell, P.G.C., Bisson, M., Sequential extraction procedure for the speciation of particulate trace elements, *Anal. Chem.*, Vol. 51, 1979, pp. 844–851.

USEPA, 1994. 600/R-94-111 reference methodology method 200.2: sample preparation. Procedure for Spectrochemical Analyses of Total Recoverable Elements.

Yu, R.L., Yuan, X., Zhao, Y.H., Hu, G.R., Tu, X.L., Heavy metal pollution in intertidal sediments from Quanzhou Bay, China, *J. Environ. Sci.,* Vol. 20, 2008, pp. 664–669.

Yu, X.J., Yan, Y., Wang, W.X., The distribution and speciation of trace metals in surface sediments from the Pearl River Estuary and the Daya Bay, Southern China, *Mar. Pollut. Bull.,* Vol. 60, 2010, pp. 1364–1371.

Zhang, Y.H., Du, J. M., Background values of pollutants in sediments of the South China Sea, *Acta Oceanol. Sinica,* Vol. 27, No. 4, 2005, pp. 161–166.

Zhang, W.G., Feng, H., Chang, J.N., Qu, J.G., Heavy metal contamination in surface sediments of Yangtze River intertidal zone: An assessment from different indexes, *Environ. Pollut.,* Vol. 157, 2009, pp.1533–1543.

# Heavy Metals at a Tailing Reservoir of Sulfide Ores

YONGHONG ZHAO[1,*], TAO ZHANG[2] and XIANXIONG CHENG[2]

[1]*Jiangxi Key Laboratory of Mining & Metallurgy Environmental Pollution Control, Jiangxi University of Science and Technology, Ganzhou, P.R. China, 341000*
[2]*Faculty of Architectural and Surveying&Mapping Engineering, Jiangxi University of Science and Technology, Ganzhou, P.R. China, 341000*

**ABSTRACT:** The tailings, which are small in particle size and exposed to natural environment, especially the extremely tiny weathered secondary mineral kernel, can easily react with air, water and living things. Up to now, the reactive tailings have caused a series of environmental problems for water and soil. In this paper, physical and chemical properties of the sulfide ore tailings were analysed. Leaching experiments in laboratory were carried out and the results showed that acidic condition can accelerate the oxidation and dissolution processes of the sulfide tailings, lead to more surounding releasion and migration of heavy metals from the tailings reservoir.

## INTRODUCTION

**MINING INDUSTRY** is usually one of the most important industrial departments in developing countries, for example in China. Mining industry can rapidly promote local economic development. However, it often leads to various environmental pollution problems at the same time. The most common and serious environmental problem is heavy metals pollution from piled tailings [1].

The tailings produced from the process of ores beneficiation are piled up at tailings reservoir near the mines [2]. Generally, the tailings are small in size and exposed to the natural environment. Therefore, they are still highly reactive in surroundings, especially extremely tiny secondary mineral particles generated by weathering, which can easily react with air, water, soil and organisms nearby [3–5]. By effects of air oxidation, biological oxidation and rain leaching, acid mining drainage (AMD) contained a large amount of heavy metals will be generated [6–7]. The pollution of AMD can migrate with runoff of surface water and underground water. Then the water and soil near the mines, even the entire ecosystem will be affected by the AMD [8–9]. Weathered sulfide mineral can lead to obvious increase of heavy metal ions concentration and acidification in tailings leachate [10–12]. Some researchers have carried out field monitor for heavy metal ions in several tailings reservoir and proved that heavy metal ions could dissolve in water and migrate with the water [13–15]. Besides this, some investigators have explored the emission rules of heavy metals ions from the tailings by the experimental simulation methods [16–17].

However, the dissolution and migration processes of heavy metals in tailings reservoir are inherently complex. It typically includes dissolution, reaction, diffusion and absorption processes. Moreover, there are many factors which can influence these processes. As far as the open-air piled tailings concerned, the influential factors can be classified into two categories. One is the natural characteristics of specific tailings, such as mineral constituents, particle size, shape and packing density; the other is environmental conditions outside, such as temperature, pH value, thickness of the mineral layer and depth of the immersed water *et al.* [18]. The detailed mechanisms and systematic works about heavy metal pollution in tailings is not enough so far.

The object of this study is the specific sulfide ore tailings produced from copper mine dressing process. The purpose of this study is to analyze the chemical and physical characteristics of the sulfide ore tailings in a tailing reservoir located in Jiangxi Province of China, to illuminate the relationship of tailing characteristics with the heavy metal leaching ability, to investigate the influence of solution pH value on heavy metal leaching.

---

*Author to whom correspondence should be addressed.
E-mail: zh5552@mail.jxust.edu.cn, Tel: +86-797-8312559, Fax: +86-797-8312559

1544-8053/14/04 131-05

***Table 1. Analysis of Major Elements in Tailings.***

| Element | Cu | Zn | Pb | S | As | Cd | Sn | Fe | $SiO_2$ | CaO | MgO | $Al_2O_3$ |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Content (%) | 0.08 | 0.62 | 0.37 | 15.78 | 0.5 | 0.02 | 0.11 | 20.07 | 12.62 | 15.69 | 7.69 | 2.58 |

## MATERIALS AND METHODS

### Samples

The tailings were sampled at two vertical directions in the tailing reservoir region which are parallel and perpendicular to the release direction of the tailings. All the tailing samples were kept in polyethylene plastic bags and prepared to be analysed in laboratory.

### Analysis Method

Dry the tailing samples in oven at the temperature of 40°C, and grind them into 100 items. Grinded samples were dealt according to the following procedures: take 0.1 g sample and put it into polytetrafluoroethylene plastic crucible, add 5 ml $HNO_3$ (65%,W/W), 10 ml HF (40%,W/W), and 12 ml $HClO_4$ (70%,W/W), heat it at a low temperature until the white smoke disperses. After cooling, add 10 ml 1:1 HCl (V/V) and transfer it into 50 ml flask after dissolution [19].

The contents of Cu, Pb, Zn and Cd were analysed by ICP-AES (Baird, USA) [20]. Typical test conditions include: Rf generator frequency 40.68 MHz, input power 1100 W, cooling gas flow rate 10 $L \cdot min^{-1}$, carrier gas flow rate 0.3 $L \cdot min^{-1}$, plasma gas flow rate 0.4 $L \cdot min^{-1}$, integral time 5 s and integral numbers three times.

## RESULTS AND DISCUSSION

### Chemical Characteristics of Tailings

The chemical characteristics analysis for major elements in tailings are shown in Table 1. It can be seen from the table that poisonous heavy metals of Pb, As and Cd are present in the tailings. The other heavy metals may also be present in the AMD and their concentration may exceed the environmental safety standard.

### Release Process Simulation of Heavy Metals

*Influence of pH on Heavy Metal Release*

Pack the sulfide ore tailings of the same weight (500 g) in the same plexiglass columns (Inner Diameter 20 mm, Length 1 m). Each column was leached daily with solution of 500 ml which was controlled at a constant pH and this procedure was lasted for 50 days successively. Different columns were leached with solutions with differnet pH values which are 2, 4, 6 and 7 respectively. The concentrations of $Pb^{2+}$, $Zn^{2+}$, $Cu^{2+}$ and $Cd^{2+}$ in outflow leaching solution were tested every day. The total amount of dissolved heavy metals in all the 50 days was summed up by days for each kind of leaching solution at different pH values. The results are shown in Table 2.

Table 2 shows that increasing the acidity of leaching solutions can significantly increase the dissolution amount of heavy metals (Pb, Zn, Cu and Cd) in tailing ores. For different leaching solutions at pH 2, 4, 6 and 7, the obvious difference of heavy metal dissolution mass can be observed. Among them, the pH value of the leaching solution influence the dissolution of Cu the most remarkable. In another words, that variation of total dissolution mass of Cu in different leaching solutions is the greatest, followed by Pb, Zn and Cd. So among the four heavy metals, their dissolution sensitive degree versus the pH variation could be arrayed as Cu > Pb > Zn > Cd. The reasons of the dissolution mass variation for the four heavy metals may be complex. It can be analysed to be inhomogeneous distribution of different heavy metals in tailings, different reaction kinetic constants or balance constants of complexation for heavy metals, and different dissolution ability in acid condition. However, the detailed reasons still need more accurate studies.

The concentration evolutions of outflow leaching solutions for $Pb^{2+}$, $Zn^{2+}$,$Cu^{2+}$ and $Cd^{2+}$ when leaching solutions at different pH are used in all the 50 days are

***Table 2. Total Amount of Heavy Metals Leached from Tailings.***

| Element | pH = 2 | pH = 4 | pH = 6 | pH = 7 | Standard Deviation | Variation, % |
|---|---|---|---|---|---|---|
| Pb | 9.41 | 7.54 | 5.24 | 3.88 | 2.47 | 38.42 |
| Zn | 410.21 | 342.5 | 282.3 | 232.6 | 75.68 | 24.55 |
| Cu | 1.82 | 1.05 | 0.82 | 0.46 | 0.55 | 56.31 |
| Cd | 0.45 | 0.36 | 0.35 | 0.32 | 0.068 | 19.66 |

*Note*: The unit is mg.

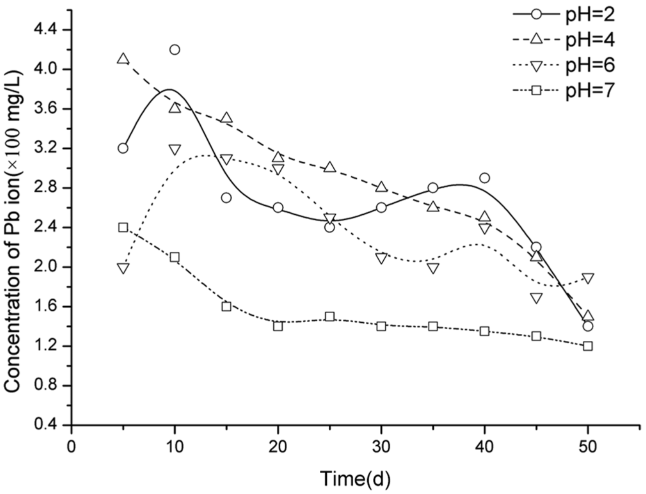


***Figure 1.*** *Concentration of Pb under different pH conditions.*

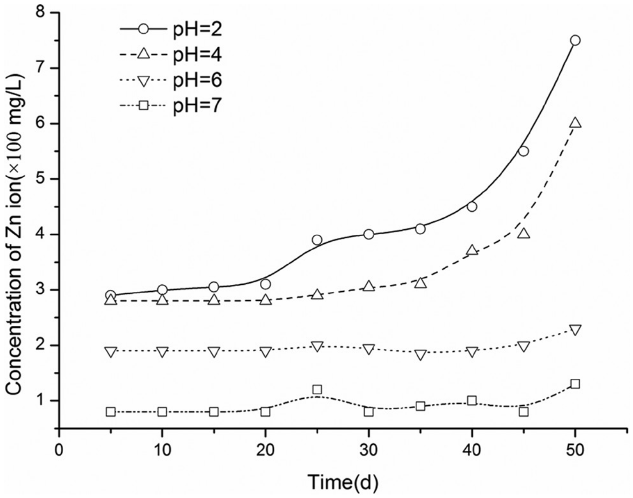


***Figure 3.*** *Concentration of Zn under differen pH conditions.*

summarized and the results are shown as Figures 1–4.

From Figure1–4, it can be known that the pH value of the leaching solution greatly influence the release of $Pb^{2+}$, $Zn^{2+}$, $Cu^{2+}$ and $Cd^{2+}$. When the pH value is less than 7, the ion concentration of tailings rises rapidly with the decreasing of the pH value. When the solution pH value is close to 7, the ion concentration of tailings changes slightly with the change of pH value. In the 50 d leaching experiment, the concentrations of $Pb^{2+}$, $Cu^{2+}$ and $Cd^{2+}$ decrease with the leaching time. Although the same leaching solution is used in 50 d for each test, the total quantity of each heavy metal in tailings is reduced with the dissolution of heavy metals. It also means the concentration of reactant is decreasing, so the dissolution mass of heavy metals decreases with the time. It is noted that the dissolution rule of $Zn^{2+}$ with the time is some especial. When the leaching solution is at a weak acid condition, the leaching time has almost no influence upon the dissolution. However, when the leaching solution is at strong acid condition, the concentration of $Zn^{2+}$ rapidly increases after the leaching of 30 d. The exact reason for this phenomenon can not be drawn out through this study. Therefore, the acidification of tailing is the critical reason for the release of heavy metal ions. In China, especially in southern China, acid rain is a very serious problem and it make the sulfide minerals to encounter a more acidification potential. These two functions will speed up the dissolution of heavy metal ions such as lead and zinc in tailings of sulfide ore. Therefore, controlling the acidification process of the tailings is an important way to reduce the environmental harm of heavy metal [21].

### *Conductivity Variation of Leaching Solutions*

Leaching solutions of every 5 d were taken to anal-

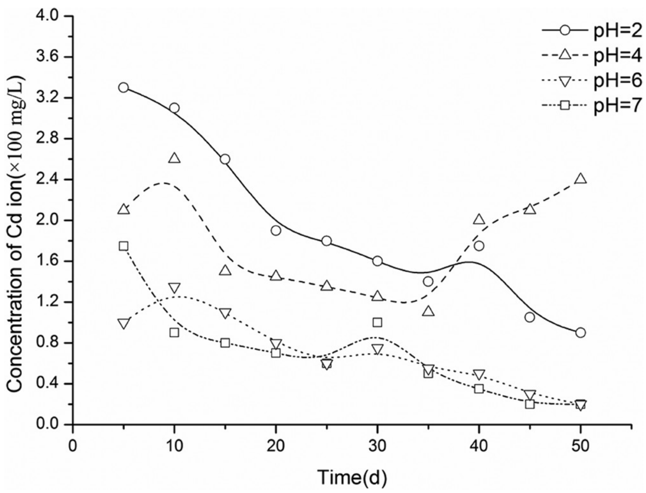


***Figure 2.*** *Concentration of Cd under different pH conditions.*

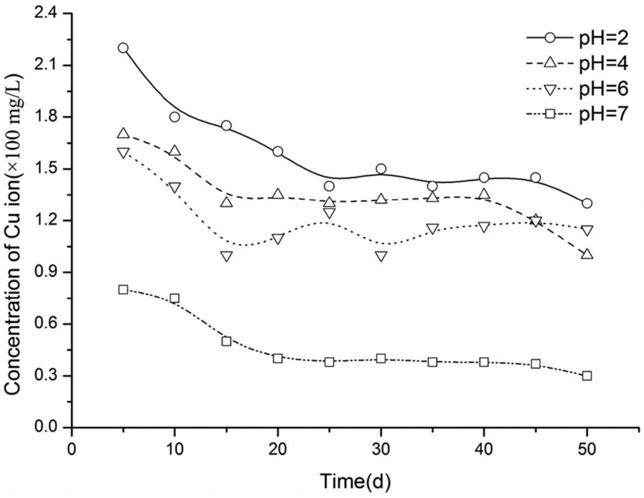


***Figure 4.*** *Concentration of Cu under different pH conditions.*

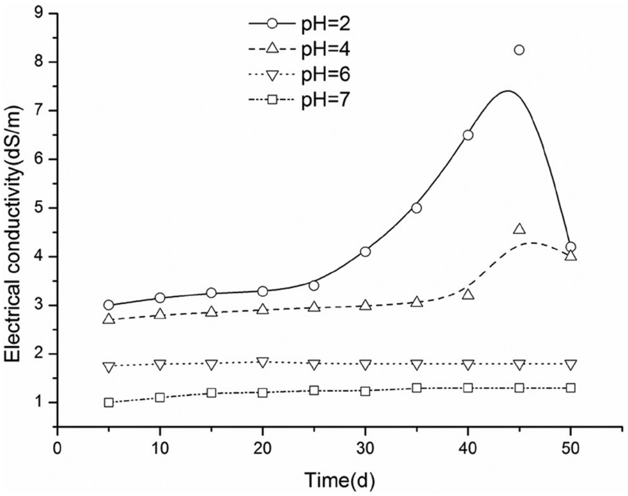


***Figure 5.*** *Conductivity variation versus time for leaching solutions.*

yse the electrical conductivity (EC). The EC analysis results are shown in Figure 5. It can be seen that when pH value is less than 4, the conductivity of leaching solutions increases significantly after leaching for 30 d. When the leaching solution is weakly acidic or weakly alkaline, the change of EC is little and the EC maintains between 0.5–1.5 ds·m$^{-1}$ during the experimental stage of 50 d. The results show that the pH value of leaching solution has direct relationship with the EC, acid condition can enhance the dissolution of solid material and lead to the increasing of salt

## CONCLUSIONS

The sulfide ore is a typical mineral in China and a large amount of sulfide ore tailings are piled up in natural environment. The tailings exposed to the natural environment may lead to a series of environmental pollution problems. Based on the analysis of physical and chemical properties of major minerals in sulfide ore tailings, it has been proved that the poisonous heavy metals (Pb, As and Cd) are present in the tailings, which may lead to a great environmental risk. Leaching experiments of the tailings show that the acid condition can enhance the dissolution of heavy metals from the tailings. Electrical conductivity test has also proved that the stronger the acid condition, the more heavy metals is dissolved in leachate. Besides that, the electrical conductivity of leachate is increasing with the time.

## ACKNOWLEDGEMENTS

The authors would like to express their gratitude to the financial support for National Natural Science Foundation of China (No. 51064007), Natural Science Foundation of Jiangxi Province (No. 20142BAB204004), External Cooperation Planned Project of Jiangxi Provincial Department of Science and Technology (No. 20111bdh80032) and Supportive Planned Project of Jiangxi Provincial Department of Science and Technology (No. 20123BBF60170).

## REFERENCES

1. Rodríguez L, Ruiz E, Alonso-Azcárate J, *et al.* “Heavy metal distribution and chemical speciation in tailings and soils around a Pb–Zn mine in Spain.” *Journal of Environmental Management 90*(2009): 1106–1116.
2. Fu J Y. “Migration regulation of heavy metals tailings of Pangushan tungsten ore.” *Jiangxi Nonferrous Metals 2*(2012):87–90.
3. Chu N. “Environmental Effect of Heavy Metal Elements in Dexing Copper MineJiangxi Province.” *Acta Geologica Sinica 04*(2008):563–68.
4. Huang K J. “Environmental geochemistry of heavy metal and trace elements in tailings of Tongliishan Copper and Iron Mine, Daye, Hubei Province.” *Geochimica 03*(2008): 213–16.
5. Wang S H. “Environmental Effects of Heavy Metal Elements Release in Yangshanchong Tailing Pool, Shizishan, Tongling, Anhui Province.” *Geological Journal of China Universities 01*(2011):93–96.
6. Akcil A, Koldas S. “Acid Mine Drainage (AMD): causes, treatment and case studies.” *Journal of Cleaner Production 14*(2006): 1139–1145.
7. Schippers A, Jozsa P G, Sand W, *et al.* “Microbiological pyrite oxidation in a mine tailings heap and its relevance to the death of vegetation.” *Geomicrobiology Journal 17*(2000): 151–162.
8. Luan HL. “Relationship of Complex Pollution to Heavy Metals Release and Migration from Floatation Tailings Dam.” *Nonferrous Metals 04*(2006):66–70.
9. Zhou Y. “Migration of heavy metals in Yangshanchong tailings impoundment in Tongling, Anhui Province.” *Res Environ Sci 4*(2010):497–99.
10. Bao R X. “A Modern Numerical Simulation Method for Heavy metal Pollutants Migration of Tailings.” *China Safety Sci J 12*(2010):39–42.
11. Brown G E. “Mineral surfaces and bioavailability of heavy metals: A molecular-scale perspective.” *Proceedings of the National Academy of Sciences 96*(1999): 3388–95.
12. Zhu J B. “Heavy metal geochemistry behavior during the oxidation of the Fankou Pb-Zn mine tailings in Guangdong province and the implications for environmental remediation of the mine.” *Acta Scientiae Circumstantiae 03* (2005): 415–41.
13. Manzano M. “The impact of the Aznalcollar mine tailing spill on groundwater.” *Science of the total environment 242*(1999): 189–209.
14. Mian M H. “Tailings erosion and resuspension in two mine tailings ponds due to wind waves.” *Advances in Environmental Research 7*(2003): 745–65.
15. Azcue J M. “Impact of abandoned mine tailings on the arsenic concentrations in Moira Lake, Ontario.” *Journal of Geochemical Exploration 52*(1995): 81–89.
16. Papassiopi N. “Removal of heavy metals from calcareous contaminated soils by EDTA leaching.” *Water, Air and Soil Pollution 109*(1999): 1–15.
17. Dijkstra JJ. “Leaching of heavy metals from contaminated soils:an experimental and modeling study.” Environ Sci Tech 38(2004): 4390–95.
18. Lin Meiqun.“Simulation Study on Influence of Environmental Factors on Release of Heavy Metal Ions out of Sulfide Ore Tailings.” *Metal mine, 384*(2008):108–11.
19. Motsi T. “Adsorption of heavy metals from acid mine drainage by natural zeolite.” *Int J Mineral Proc 92*(2009): 42–48.

20. Rios C A.”Removal of heavy metals from acid mine drainage (AMD) using coal fly ash, natural clinker and synthetic zeolites.” *J Hazard. Mat. 156*(2008): 23–35.

21. Li L. "Antimony and arsenic migration and environmental impacts on river draining in the Dachang multi-metalliferous mine area in Guangxi, China.” *Res Environ Sci 6*(2009):682–86.

# Aqueous Cr(VI) Adsorption to Sludge using a Marble Processing Wastewater Treatment Method

MERVE SOGANCIOGLU* and ESRA YEL

*Selcuk University, Engineering Faculty, Environmental Engineering Department, Konya, Turkey*

**ABSTRACT:** Cr(VI) adsorption onto dried marble processig wastewater (non-treated wastewater) solids (MPW) and dried physicochemical marble sludge from coagulation-flocculation treatment (MPWT) of an andesite processing plant's wastewater was investigated. MPW sludge and MPWT sludge were incorpotared as inorganic adsorbents in batch reactors containing aqueous Cr(VI) solutions varying between 1 and 25 mg/L. MPWT sludge was obtained with alum coagulant. A dry particulate (powder) form of sludge samples were used at a 5 g/100 mL dosage. Isotherm studies indicated that Cr(VI) adsorption by these andesite- oriented adsorbents may be characterized using a Freundlich model for MPW sludge and a Langmuir model for MPWT sludge.

## INTRODUCTION

**ANDESITE,** a type of marble, is a heat and frost resistant volcanic rock commonly used in architecture, building, and art since ancient times. Large-scale andesite production has generated a considerable amount of waste materials. almost 70% of this gets wasted in the mining, processing, and polishing stages. These wastes have a serious impact on the environment since they contain non-degradable, diverse-sized marble particles. Direct discharge of andesite processing wastewaters into the environment will have considerable adverse effects due to inert andesite colloids content. The most common method for treatment is coagulation and/or flocculation via common coagulants or flocculants. However, the physicochemical treatment of andesite processing wastewater is more difficult than for other types of marble. The treatment of a large volume of andesite processing wastewater will generate large quantities of physico-chemical sludges. Different coagulants and flocculants have been studied for andesite processing wastewater and the physico-chemical sludges from these have different characteristics [1]. Land disposal of these physicochemical sludges has adverse effects on the environment. There are also potential problems due to large areas of land that are required. Therefore, final disposal alternatives other than land disposal need to be investigated.

Industrial wastewaters are frequently laden with toxic heavy metals in which significant amounts are deposited into natural aquatic and terrestrial ecosystems [2]. Among these heavy metals, chromium (Cr) is one of the pollutants in surface water and groundwater resulting from numerous industrial activities such as preservation of wood, textile dyeing, leather tanning, electroplating, and metal finishing [3]. In general, chromium exists in the trivalent Cr(III) and hexavalent Cr(VI) states. Chromium (VI) is more toxic than the Cr(III) form. Because of its high toxicity, Cr(VI) must be removed from wastewater before being discharged to an aquatic system. Common Cr (VI) anions, chromate ($CrO_4^{2-}$) and dichromate ($Cr_2O_7^{2-}$), are strong oxidants, and chromate is a known carcinogen and a suspected mutagen and teratogen in biological systems [4]. Cr(VI) concentration in various industrial effluents has been reported over a wide range from 35–555 mg/L Cr(VI) [5].

Adsorption is a heavy metal removal method for aqueous systems. As a common adsorbent, activated carbon has been used in treatment but it is ineffective on many metals. Sometimes its high cost restricts its use. Cheaper and effective adsorbents and many non-conventional low-cost adsorbents such as clay materials, zeolites, siliceous material, agricultural wastes, and industrial waste products have also been suggested [6].

---

*Author to whom correspondence should be addressed.
E-mail: merves@selcuk.edu.tr

1544-8053/14/04 137-05

In this study, dried raw marble processing wastewater (MPW) sludge and dried marble processing wastewater treatment (MPWT) sludge originated from coagulation-flocculation treatment of an andesite processing plant wastewater as two different adsorbents. Removal of Cr (VI) from aqueous solutions using these two sludges was investigated.

## MATERIAL AND METHOD

### Materials

In andesite processing plants, raw andesite powder containing wastewater was generated as a result of wet cutting [1]. The andesite processing wastewater properties and the chemical composition of the andesite particles are given in Table 1. The wastewater total solids and suspended solids were equal and very high in concentration. This implies that no dissolved solids existed, all the solids were suspended, andesite particles were colloidal in size, and they were not settleable. MPW sludge was obtained by directly drying the raw wastewater and marble powders were used as adsorbent. The other adsorbent (MPWT sludge), physicochemical sludge, was obtained by coagulation-flocculation process with alum. Coagulation (150 rpm for 90 second), flocculation (30 rpm for 15 min), and sedimentation (30 min) steps were carried out within a Jar Test device [7]. The coagulation flocculation sludge samples were dried at room temperature in the laboratory.

Stock Cr(VI) solution (100 mg/L) was prepared by dissolving potassium dichromate ($K_2Cr_2O_7$) in distilled water. The stock solution was then diluted to obtain standard solutions (1, 5, 10, 15, 20, and 25 mg/L). Reagents used for Cr(VI) analysis were $HNO_3$, $H_2SO_4$, 1,5 diphenylcarbazide, and $K_2Cr_2O_7 \cdot 2H_2O$. All solutions were prepared with deionized water.

***Table 1. Properties of Andesite Processing Wastewater.***

| Parameter | Value |
|---|---|
| pH | 8.4–9.5 |
| Total solids, (g/L) | 25–37 |
| Ca (ppm) | 5317.64 |
| Mg (ppm) | 199.34 |
| Na (ppm) | 288.03 |
| Fe(ppm) | 13.83 |
| K (ppm) | 0.0 |
| Pb (ppm) | 0.16 |
| Cd (ppm) | 0.25 |
| Zn (ppm) | 0.57 |
| $H_2CO_3$(ppm) | 39.0 |
| Cr(ppm) | 0.0 |

### Batch Adsorption Experiments

Reactors of 100 ml liquid volume and 5 g adsorbent were operated in the shaker (150 rpm) until equilibrium and change of concentration against time was observed. Preliminary trials indicated that 120 minutes was sufficient to achieve equilibrium. Initial Cr (VI) concentrations in the reactors were varied from 1 mg/L to 25 mg/L, respectively, at 25°C and pH 7. Samples were filtered and the Cr(VI) that remained in the filtrate was analyzed.

### Analysis

Cr(VI) were determined according to Standard Methods [8]. This procedure measures only Cr(VI) by reaction with 1,5 diphenylcarbazide in acid solution. A red-violet complex is formed and absorbance measured spectrophotometrically at 540 nm was converted to concentration using the following calibration equation $y = 0.132x$. Adsorption capacities were calculated according to Equation (1).

$$q = \frac{V(C_o - C_e)}{m} \tag{1}$$

$V$ is solution volume ($L$), $m$ is amount of adsorbent (g), and $C_o$ and $C_e$ are initial and equilibrium Cr(VI) concentrations (mg/L), respectively.

Various theoretical models could be applied to experimental data in order to find a model which adequately describes equilibrium data. Langmuir and Freundlich isotherms have been extensively used to describe the equilibrium established between adsorbed metal ions on the adsorbent ($q_e$) and metal ions remaining in solution ($C_e$) at equilibrium and at constant temperature [9]. The Langmuir model assumes a monolayer adsorption onto a surface and is represented by Equation (2) [10], The Freundlich isotherm [Equation (3)] assumes that the uptake of metal ions occurs on a heterogeneous surface by a multilayer adsorption [11],

$$q_e = \frac{q_{\max} \cdot b \cdot C_e}{(1 + b \cdot C_e)} \tag{2}$$

$$q_e = k \cdot C_e^{1/n} \tag{3}$$

where $q_{\max}$ (mg/g) is maximum adsorption capacity

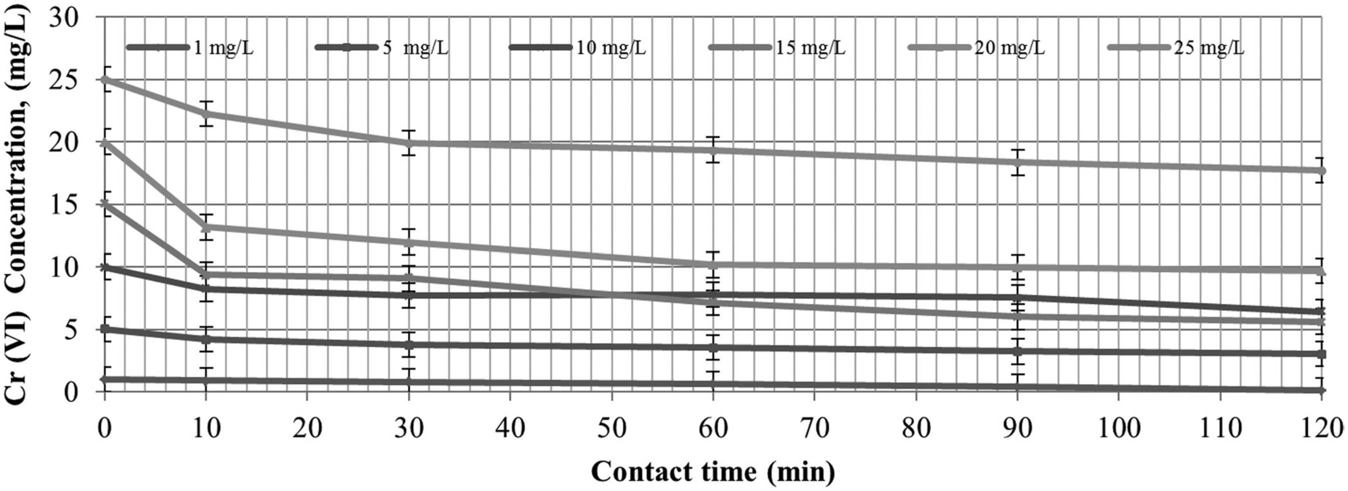


***Figure 1.*** *Effect of contact time on Cr(VI) adsorption for MPW sludge.*

and $b$ (L/g) is the Langmuir adsorption rate constant. The parameters $k$ and $n$ are Freundlich constants.

## RESULTS AND DISCUSSION

### Effect of Contact Time on Cr(VI) Adsorption

The effect of contact time on Cr(VI) concentration is presented in Figure 1 and Figure 2 for MPW sludge and MPWT sludge, respectively. The adsorption of Cr(VI) was rapid in the first 10 min and then the adsorption rate decreased for the MPW sludge. For a 25 mg/L initial Cr(VI) concentration the Cr(VI) removal performance was better than for other concentrations at 30 min (see Figure 1).

The adsorption process was similarly rapid during the first 10 min for MPWT sludge (especially for 15 mg/L Cr(VI) concentration) and then remained constant with contact time (see Figure 2). Rapid initial adsorption could be due to the fact that a large number of active sites are available for adsorption of Cr(VI) on the marble sludge. After rapid diffusion of Cr(VI) molecules from the bulk solution to the exterior surface of adsorbent particles the removal rate slowed down gradually until equilibrium.

According to Figure 1 and Figure 2, ultimate, Cr(VI) adsorption ratios of the MPW sludge and the MPWT sludge were 93% and 92 %, respectively, for an initial Cr(VI) concentration of 1 mg/L. On the other hand, these values were 29%and 35% for a 25 mg/L initial Cr(VI) concentration (Figure 3). Because of the porous morphology of marble sludge [12], as the adsorbed Cr(VI) incresed marble sludge surface area decreased. Also, adsorption efficiencies decreased for all doses. Adsorption capacities of the MPWT sludge were higher than for the MPW sludge which was closely related

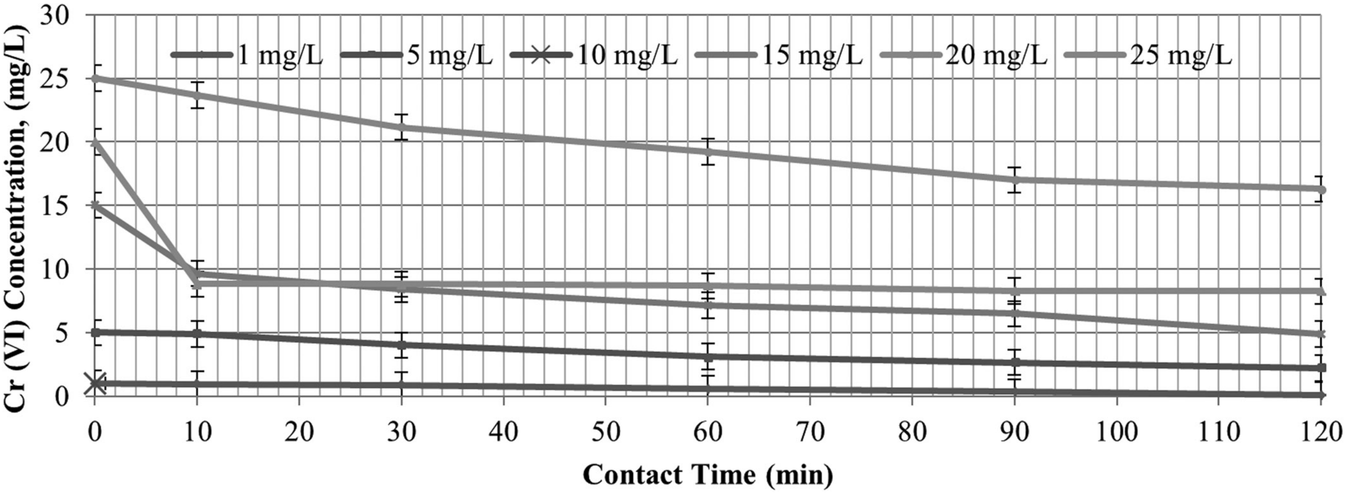


***Figure 2.*** *Effect of contact time on Cr(VI) adsorption for MPWT sludge.*

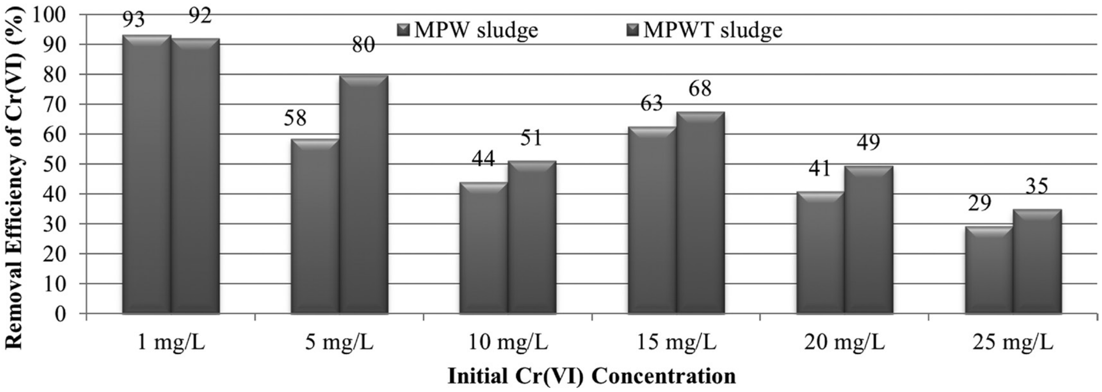
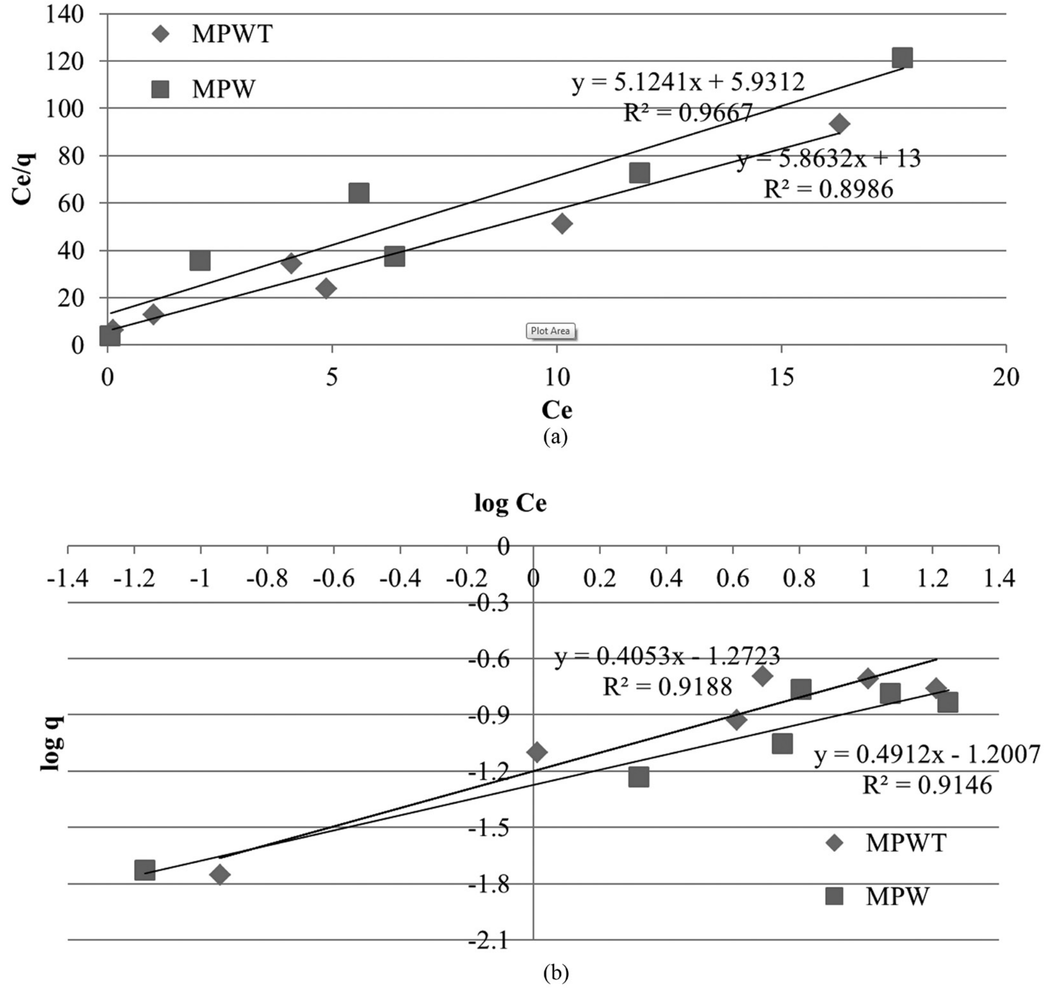


***Figure 3.*** *Removal efficiency of Cr(VI) for MPW sludge and MPWT sludge.*

to number of pores forming for both the surface and interior of the MPW sludge.

## Cr (VI) Adsorption Isotherms

Figure 4 presents the Langmuir and Freundhlich isotherms of Cr(VI) adsorption on the MPW and the MPWT. Langmuir and Freundlich constants are summarized in Table 2. The Freundlich isotherm represented a better model for MPW sludge than the Freundlich isotherm with a higher $R^2$. The surface of the MPW sludge is likely made up of small heterogeneous adsorption patches which are dissimilar to each other in respect to adsorption process. This supports the Freun-



(a)

(b)

***Figure 4.*** *(a) Cr (VI) adsorption isotherms with results fitted to the Langmuir equation and (b) Freundhlich equation.*

***Table 2. Langmuir and Freundlich Constants for Cr(VI) Adsorption.***

| | Langmuir | | | Freundlich | | |
|---|---|---|---|---|---|---|
| **Adsorbent** | $q_{max}$ (mg/g) | $b$ (1/mg) | $R^2$ | $k$ | $1/n$ | $R^2$ |
| MPW sludge | 0.76 | 2.244 | 0898 | 2.540 | 1.272 | 0.918 |
| MPWT sludge | 1.68 | 1.161 | 0.996 | 3.097 | 1.200 | 0.914 |

dlich isotherm which represents physical multilayer adsorption onto the heteregenous surface of the MPW sludge.

On the other hand, by applying the Langmuir isotherm by plotting $C_e/q_e$ against $C_e$ a straight line was obtained with high determination coefficients ($R^2 > 0.9$) confirming that the Langmuir isotherm is applicable for adsorption of Cr(VI) onto MPWT sludge. From the slope and intercept adsorption equilibrium constant, $b = 1.161$ l/mg and the maximum adsorption capacity $q_{max} = 1.680$ mg/g. Homogeneous surface of treatment sludge resulted in monolayer chemisorption onto MPWT sludge.

The adsorption maximum capacities were given in Figure 5. According to this graphic, MPWT sludge capacities were significantly higher than MPW sludge capacities. Because that effect of used coagulant, flocs were occurred homogenious adsorbent surface. The Cr(VI) adsorption was strongly Cr(VI) concentration dependent. The increase in the Cr(VI) concentration enhances the interactions between Cr(VI) and physicochemical sludge particles. Therefore, the amount adsorbed decreased with higher initial Cr(VI) concentrations for MPWT sludge.

## CONCLUSION

In this study, adsorption processes are characterized using a Freundlich model for MPW sludge and a Langmuir model for MPWT sludge with max adsorption capacities of 0.760 mg/g and 1.680 mg/g, respectively.

Results indicated that MPWT sludge can be an adsorbent for removal of Cr(VI) from aqueous solutions. Attractive features of coagulation and flocculation sludge are that it is environmentally friendly, effective, and low cost. Moreover, when this sludge has further utilization land requirements decrease. Therefore, this application helps preserve land and sustainability concepts. As further development occurs, temperature and pH effects may be investigated to increase system performance.

## REFRENCES

1. Soganciogluu M., Yel E., Keskin U.S., Utilization of andesite processing wastewater treatment sludge as admixture in concrete mix, *Construction and Building Materials, 46* (2013) 150–155.
2. J.P. Chen, L.L. Lim, Key factors in chemical reduction by hydrazine for recovery of precious metals, *Chemosphere 49* (2002) 363–370.
3. S.D. Kim, K.S. Park, M.B. Gu, Toxicity of hexavalent chromium to Daphnia magna: influence of reduction reaction by ferrous iron, *J. Hazard. Mater. A93* (2002) 155–164.
4. D.E. Cummings, S. Fendorf, N. Singh, B.M. Peyton, T.S. Magnuson, Reduction of Cr(VI) under acidic conditions by the facultative Fe(III)-reducing bacterium Acidiphilium cryptum, *Environ. Sci. Technol. 41* (2007) 146–152.
5. Arceivala J. Soli, *Industrial Wastewater Treatment for Reuse and Removal,* Middle Technical University, Enviromental Engineering Department Publications. (1976) 315.
6. R.S. Juang, F.C. Wu, R.L. Tseng, *Environ. Technol. 18* (1997) 525.
7. Yilmazturk D. Physico-chemical treatment of andesite processing wastewater. MS thesis. Konya, Selcuk University, 2011.
8. Standard Methods for the Examination of Water & Wastewater, American Public Health Association, 2005.
9. Garg, U.K., Kaur, M.P., Garg, V.K., Sud, D.,Removal of hexavalent chromium from aqueous solution by agricultural waste biomass. *J. Hazard. Mater. 140* (2007) 60–68.
10. The constitution and fundamental properties of solids and liquids. Irving Langmuir; J. Am. Chem. Soc. 38(1916), 2221–2295.
11. Freundlich HMF. Uber die adsorption in losungen. *Z Phys Chem.* (1906) 57 385–470.
12. Hamed ,Mostafa M., Ahmed, Metwally, I.M., S.S. , Adsorptive removal of methylene blue as organic pollutant by marble dust as eco-friendly sorbent, *Journal of Industrial and Engineering Chemistry* (2013).

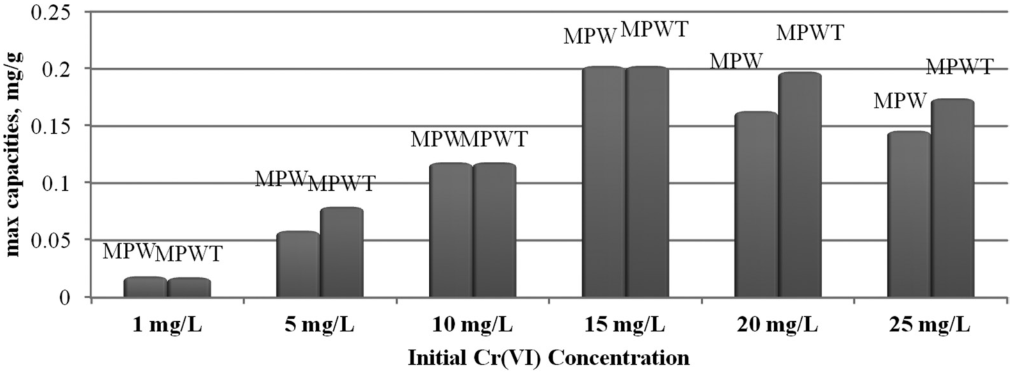


***Figure 5.*** *Adsorption capacities.*

# GUIDE TO AUTHORS

1. Manuscripts shall be sent electronically to the Editor-in-Chief, Dr. P. Brent Duncan at pduncan@unt.edu using Microsoft Word in an IBM/PC format. If electronic submission is not possible, three paper copies of double-spaced manuscripts may be sent to Dr. P. Brent Duncan, (Editor of the *Journal of Residuals Science & Technology,* University of North Texas, Biology Building, Rm 210, 1510 Chestnut St., Denton, TX 76203-5017) (Tel: 940-565-4350). Manuscripts should normally be limited to the space equivalent of 6,000 words. The editormay waive this requirement in special occasions. As a guideline, each page of a double-spaced manuscript contains about 300 words. Include on the title page the names, affiliations, and addresses of all the authors, and identify one author as the corresponding author. Because communication between the editor and the authors will be electronic, the email address of the corresponding author is required. Papers under review, accepted for publication, or published elsewhere in journals are normally not accepted for publication in the *Journal of Residuals Science & Technology*. Papers published as proceedings of conferences are welcomed.
2. Article titles should be brief, followed by the author's name(s), affiliation, address, country, and postal code (zip) of author(s). Indicate to whom correspondence and proofs should be sent, including telephone and fax numbers and e-mail address.
3. Include a 100-word or less abstract and at least six keywords.
4. If electronic art files are not supplied, submit three copies of camera-ready drawings and glossy photographs. Drawings should be uniformly sized, if possible, planned for 50% reduction. Art that is sent electronically should be saved in either a .tif or .JPEG files for superior reproduction. All illustrations of any kind must be numbered and mentioned in the text. Captions for illustrations should all be typed on a separate sheet(s) and should be understandable without reference to the text.
5. DEStech uses a numbered reference system consisting of two elements: a numbered list of all references and (in the text itself) numbers in brackets that correspond to the list. At the end of your article, please supply a numbered list of all references (books, journals, web sites etc.). References on the list should be in the form given below. In the text write the number in brackets corresponding to the reference on the list. Place the number in brackets inside the final period of the sentence cited by the reference. Here is an example [2].

   *Journal:* 1. Halpin, J. C., "article title", *J. Cellular Plastics,* Vol. 3, No. 2, 1997, pp. 432–435.

   *Book: 2.* Kececioglu, D. B. and F.-B. Sun. 2002. *Burn-In Testing: Its Quantification and Optimization,* Lancaster, PA: DEStech Publications, Inc.
6. Tables. Number consecutively and insert closest to where firstmentioned in text or type on a numbered, separate page. Please use Arabic numerals and supply a heading. Column headings should be explanatory and carry units. (See example at right.)
7. Units & Abbreviations. Metric units are preferred. English units or other equivalents should appear in parentheses if necessary.
8. Symbols. A list of symbols used and their meanings should be included.

***Table 5. Comparison of state-of-the-art matrix resins with VPSP/BMI copolymers.***

| Resin System | Core Temp. (DSC peak) | $T_E$ | Char Yield, % |
|---|---|---|---|
| Epoxy (MY720) | 235 | 250 | 30 |
| Bismaleimide (H795) | 282 | >400 | 48 |
| VPSP/Bismaleimide copolymer | | | |
| C379: H795 = 1.9 | 245 | >400 | 50 |
| C379: H795 = 1.4 | 285 | >400 | 53 |

9. Page proofs. Authors will receive page proofs by E-mail. Proof pages will be in a .PDF file, which can be read by Acrobat Reader. Corrections on proof pages should be limited to the correction of errors. Authors should print out pages that require corrections and mark the corrections on the printed pages. Pages with corrections should be returned by FAX (717-509-6100) or mail to the publisher (DEStech Publications, Inc., 439 North Duke Street, Lancaster, PA 17602, USA). If authors cannot handle proofs in a .PDF file format, please notify the Editor, Dr. P. Brent Duncan at pduncan@unt.edu.
10. Index terms.With proof pages authors will receive a form for listing key words that will appear in the index. Please fill out this form with index terms and return it.
11. Copyright Information. All original journal articles are copyrighted in the name of DEStech Publications, Inc. All original articles accepted for publication must be accompanied by a signed copyright transfer agreement available from the journal editor. Previously copyrighted material used in an article can be published with the *written* permission of the copyright holder (see #14 below).
12. Headings. Your article should be structured with unnumbered headings. Normally two headings are used as follows:

    Main Subhead: DESIGN OF A MICROWAVE INSTALLATION Secondary Subhead: Principle of the Design Method

    If further subordination is required, please limit to no more than one (*Third Subhead*).
13. Equations. Number equations with Arabic numbers enclosed in parentheses at the right-hand margin. Type superscripts and subscripts clearly above or below the baseline, or mark them with a caret. Be sure that all symbols, letters, and numbers are distinguishable (e.g., "oh" or zero, one or lowercase "el," "vee" or Greek nu).
14. Permissions. The author of a paper is responsible for obtaining releases for the use of copyrighted figures, tables, or excerpts longer than 200 words used in his/her paper. Copyright releases are permissions to reprint previously copyrighted material. Releases must be obtained from the copyright holder, which is usually a publisher. Forms for copyright release will be sent by the editor to authors on request.

**General:** The *Journal of Residuals Science & Technology* and DEStech Publications, Inc. are not responsible for the views expressed by individual contributors in articles published in the journal.