

# 54 (2015) 1353–1360 April



# Science Walden: new horizons of combined ecological sanitation with separated urine/feces and treatment wetlands

Jongkwan Park<sup>a</sup>, Kyongmi Chon<sup>b</sup>, Jaeweon Cho<sup>a,\*</sup>

<sup>a</sup>Department of Civil and Environmental Engineering, Yonsei University, 50 Yonsei-ro, Seodaemun-gu, Seoul 120-749, Korea, Tel. +82 2 2123 8296; Fax: +82 2 364 5300; email: chojw@yonsei.ac.kr

<sup>b</sup>School of Environmental Science and Engineering, GIST, 123 Cheomdan-gwagiro, Buk-gu, Gwangju 500-712, Korea

Received 25 December 2013; Accepted 26 February 2014

#### ABSTRACT

A new engineering concept, designed with separated urine/feces and graywater recovery, was proposed, and the first steps in taking this concept from the planning stage to implementation have been taken using pilot experiments within a small village. The village employs ecological toilet, equipped with 24 h ventilation, and constructed treatment wetlands consisting of both vertical and horizontal subsurface wetlands for graywater recovery. The quality of recovered graywater was similar to that of the water found in the adjacent stream, which is a concept of zero discharge system with graywater. Separated urine, in either fresh or stored form, was diluted with collected rainwater and with reused water without any detergent from a sink, to be supplied to vegetable garden as fertilizer. Separated dried feces were composted in the garden for approximately 3 months and then used as fertilizer. Dried and composted feces, together with stored urine, were characterized in terms of microbial community using pyrosequencing, to identify the presence of any potential pathogens, in order to confirm the system provides safe hygiene. Hypothesized idea of a micro-algae farm within the village might be proposed with separated urine serving as nutrients for algae that could in turn be cultivated as biofuel (diesel) produced from extracted lipids of algae. Through this pilot village test, we have taken a great stride towards practical realization of experimental concepts, in the form of this new urban water management model with ecological sanitation.

*Keywords:* Urban water management; Ecological sanitation; Treatment wetland; Urine; Feces; Water recovery

# 1. Introduction

Considering that urine and feces waste consists of nitrogen, phosphorus, carbon, and others, this charac-

\*Corresponding author.

terization might seem misapplied. However, the scale of the problem highlights why wastewater treatment is an important area in need of improvement: the city of Seoul, for example, with a population of approximately 10 million in 2012 [1], excretes approximately 12 and 31% of total nitrogen (TN) contained in the

Presented at the 6th International Conference on the "Challenges in Environmental Science and Engineering" (CESE-2013), 29 October–2 November 2013, Daegu, Korea

1944-3994/1944-3986 © 2014 Balaban Desalination Publications. All rights reserved.

influent of raw wastewater into the wastewater treatment plants, through urine (approximate 19% of N, 5% of P, 17% of C, 6% of Ca, and 53% of others, as based on dry mass) and feces (approximate 7% of N, 5% of P, 50% of C, 5% of Ca, and 33% of others, as based on dry mass), respectively. A single person excretes approximate 1.0–1.31 of urine and 135–270 g of feces in average per day [2–5].

It is not difficult to envision a much better water-quality management plan for the Han River. This river receives the discharged effluent waters from Seoul's wastewater treatment plants, and separating out these wastes from the water would be both environmentally friendly and more efficient. Our proposed plan to enact this waste separation may be only a virtual concept, but it can be executed through existing engineering concepts such as separation of urine, ecological sanitation system, and others.

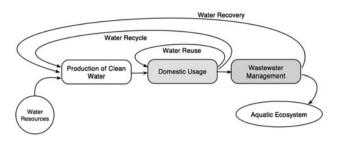


Fig. 1. Concepts of water reuse, water recycle, and water recovery.

Urine for fertilizer can be used in either a relatively fresh form or stored for a period (>6 months) [6]. After a certain period of storage, urine undergoes significant change in chemical characteristics: the major nitrogen form turns from urea to ammonia (due to urea hydrolysis), with subsequent significant

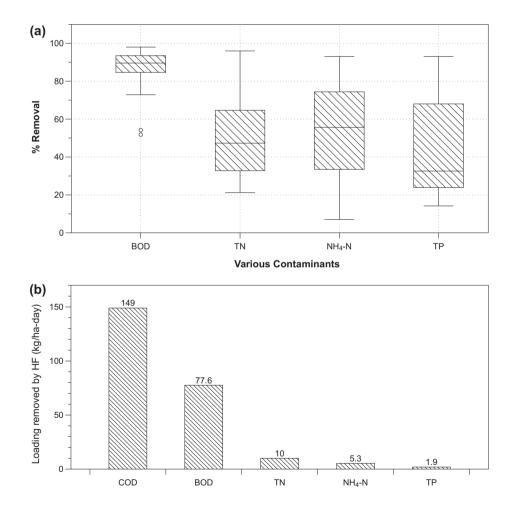


Fig. 2. Summary of (a) contaminants control and (b) average loading (based on removal) by horizontal subsurface flow treatment wetlands (drawn using data provided in the reference; data represented here exhibit high and low limits, median, and first and third quarter values) [12].

increases of pH and conductivity [7]. Treatment wetlands have been recognized as desirable methods for wastewater treatment because of low energy consumption and maintenance requirements [8]. Especially, reclamation of graywater using treatment wetland has been being studied to reduce fresh water consumption in households [9,10]. In this project, to demonstrate the possibility of applying practical engineering concepts for urine/feces separation from sanitation processes and subsequent graywater reclamation implementation, a pilot experiment with a relatively small village having 23 houses has been attempted. All houses used in the test employed an ecological toilet system equipped with a ventilation unit. With separation of urine and feces from sanitation, different concepts of reusing water can be implemented; thus, in this paper, three different terminologies regarding re-using water are introduced, namely, water reuse, water recycling, and water recovery (denoted as water 3R), as conceptually represented in Fig. 1. These distinctions are similar to the ones made in solid waste and resource recovery [11]. The act of using used water without any prior treatment is defined as water reuse. When used water is sent back to a certain treatment step, depending upon water quality, prior to being discharged to wastewater management, it is defined as water recycling. When used water is sent back to a certain treatment control step, after being discharged to wastewater management, such as sewage, it is defined as water recovery. In this paper, three different terminologies (with different meanings) of re-using water in the pilot tests are utilized.

As shown in Fig. 2, control efficiency values, taken from 17 different case studies, vary widely but show some potential of contaminants control, depending on conditions in 11 different countries [12]. After contaminant control with treatment wetlands, water is expected to be reused (as in the concept of water recovery) after identification of water qualities; in this pilot experiment with the village, recovered water was sent back to each house to be further used for gardening. Contaminants removal rates (Fig. 2(a)) and loading rates based on removed contaminants with horizontal subsurface treatment wetlands (Fig. 2(b)) are summarized. These were used as a reference to design the treatment wetlands with graywater from the pilot village in terms of required surface area of treatment wetland with graywater loading.

Nutrients contained in wastewater effluent can be used to grow micro-algae, which in turn can provide lipids to be transformed into biofuel (e.g. biodiesel) [13]. Similarly, it can be expected that urine (either fresh or stored) is used as nutrients for micro-algae cultivation, which leads to a concept (represented in the Concluding Section, in this paper) in which separated urine is not mixed with graywater but may be used in a micro-algae farm [14]. Insights on both nutrients specification (especially nitrogen (urea, ammonia, or nitrate/nitrite)) and levels of salts may be elucidated prior to being used as food for microalgae; thus, in this pilot experiment, both fresh and stored urine were characterized in terms of nitrogen speciation, and salts levels were attempted to be controlled using a nanofiltration (NF) membrane.

# 2. Material and methods

#### 2.1. Urine and feces sampling

Urine, both fresh and stored for a certain period (generally longer than 6 months), was used as fertilizer after dilution for vegetable gardens in the village. The water for dilution was collected into isolated tanks from rainwater and used water which was drained kitchen sinks (the concept of water reuse, as shown in Fig. 1). The effects of using either urine fertilizer, or fresh or stored urine on vegetation growth and other agricultural results were not demonstrated in this study, but urine fertilizer has been continuously utilized by most houses in the village. Feces were stored and dried inside the ecological toilets, and, then, transported to gardens after storing for approximately 3-6 months under ventilated conditions. Please note that a major objective of this pilot experiment and subsequent paper is to deduce a new engineering concept or idea to be practically utilized for urban water management; thus, not all the experiments in the pilot test have been thoroughly conducted with robust evaluating measurements. However, hygiene of using both urine and feces was checked, with respect to its microbial community using a genetic approach; the bacterial 16S RNA gene was used for pyrosequencing [15-18]. One liter of stored urine was taken from one of the houses where urine is used as fertilizer. Dried feces and compost were also collected from the same place. Those samples were transported to the laboratory within 24 h and then stored at 4°C until further analyses. The water quality of rainwater was monitored for one year in 2012, in terms of pH, TOC, TN, ammonium ion, and selected heavy metals, including lead. Data were not shown in this paper. Only ammonium ion levels were somewhat high (approximately>3 mgN/l (>5 mgN/l as TN)), especially in spring season compared to other seasons. However, there were no significant obstacles to using rainwater as the water component of the reusing urine schemes previously described. With urine and feces being separated, only

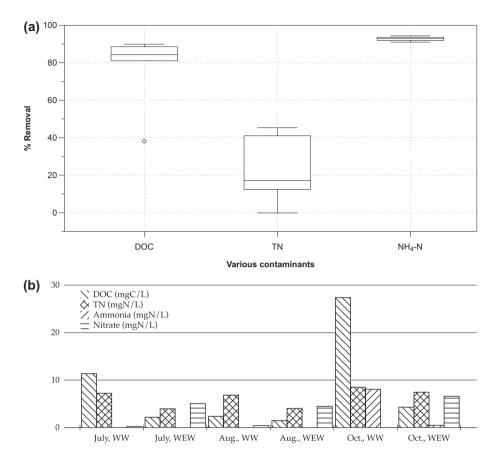


Fig. 3. Observed contaminants controls in the treatment wetland of the pilot village, with respect to DOC and nitrogen, in terms of removal efficiencies and levels in the effluents (WW: wastewater, i.e. graywater; WEW: effluent of the treatment wetland).

graywater is conveyed to the treatment wetland. With a treatment capacity of  $15 \text{ m}^3/\text{d}$ , in the pilot village; the wetland employs vertical followed by horizontal

subsurface flow types of constructed systems, as planned, designed, and being operated based on treatment efficiencies and removed loading rates obtained

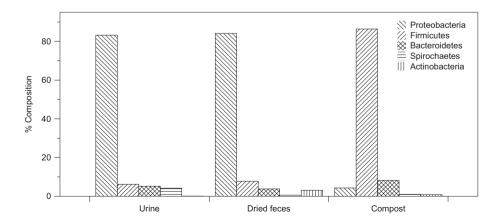


Fig. 4. Results of microbial community analyses, using pyrosequencing, in phylum levels, with stored urine, dried feces after approximately 1 month storage under ventilation, and compost with dried feces mixed with gardening soils after at least 3 months composting.

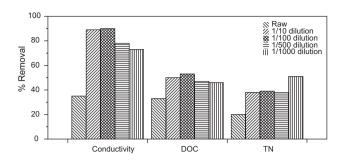


Fig. 5. Percentage removal patterns of raw and diluted urine samples, with respect to conductivity, DOC, and TN.

from the published results, as depicted in Fig. 2(a) and (b) [12].

#### 2.2. Water quality analysis

The water qualities of urine, graywater, and reclaimed graywater through treatment wetland were analyzed. Levels of dissolved organic carbon (DOC) and TN were measured using a non-purgeable method with an organic analyzer (TOC-V<sub>CPH</sub>, TNM-1 Shimadzu, Kyoto, Japan). Ammonium, nitrate, and nitrite levels of samples were measured using an ion chromatography apparatus (DX-120, ICS-90, ICS-900, Dionex, CA, USA), equipped with an IonPac columns (AS14 and CS12A, Dionex, CA, USA). Due to concentration of various ions and DOC contained in urine, too high to be measured, the samples were diluted prior to measurements (1,000-fold and 200-fold dilutions with organic carbon and other ions analyses).

# 2.3. Pyrosequencing

With microbial community analysis using pyrosequencing, the total bacteria genomic DNA was extracted using a Fast DNA SPIN Kit according to the manufacturer's instructions. The extracted DNA was amplified using suitable forward and reverse primers targeting the V1 to V3 regions of the bacterial 16S rRNA gene. PCR reaction was performed under the following conditions: initial denaturation at 95°C for 5 min, followed by 30 cycles of denaturation at 95°C for 30 s, annealing at 55°C at for 30 s, and extension at 72°C for 30s; followed by final elongation at 72°C for 7 min. The amplified products were purified using a PCR purification kit (Qiagen, Valencia, CA, USA), 1 µg of each amplified product was mixed and subjected to DNA pyrosequencing. The pyrosequencing was performed at the Chun Lab, Inc. (Seoul, Korea) with a 454 Genome Sequencer FLX Titanium (Roche, Basel, Swiss). Sequence reads from samples were separated by unique barcodes. Then, the barcodes, linker, and PCR primer sequences were removed from the original sequencing reads. The trimmed sequence reads were subjected to a filtering process where only reads containing 0-1 ambiguous bases (Ns) and more than 300 base pairs were selected for the final bioinformatics analyses. For the taxonomic assignment of each pyrosequencing read, the EzTaxon-extended database -(www.eztaxon-e.org) was used, which provides 16S rRNA gene sequences of type strains that have valid published names and representative phylo types of either cultured or uncultured entries in the Gen Bank public database with complete hierarchical taxonomic classification from phylum to species.

# 2.4. Salt removal experiment

For a conceptual design of a micro-algae cultivating farm, certain levels of salts might be one of several limiting factors for culturing, depending upon algae species. Filtration experiments using a NF membrane with a molecular weight cutoff (MWCO) of 250 Da (membrane code NE70 from the Woongjin Chemical, Korea) were attempted to reduce conductivity level, with other nutrients, such as TN, being maintained to the highest level. Besides a nominal MWCO value provided by the manufacturer, the MWCO was identified using the fractional rejection method developed in a previous paper [19]. The tested NF (polyamide thin-film composite) membrane exhibits negative surface charges at the pH range of 4-10, as measured using the electrophoresis method with non-charged nanoparticles [20]. Various urine samples were used for the filtration, including raw (with conductivity, DOC, and TN of 23,580 µS/cm, 4,591 mgC/l, and

Table 1 The result of NF filtration depending on the urine dilution rate

Dilution rate	Conductivity (µS/cm)	DOC (mgC/l)	TN (mgN/l)
10	349	204	362
100	29	19	35
500	13	4	7
1,000	7	3	4

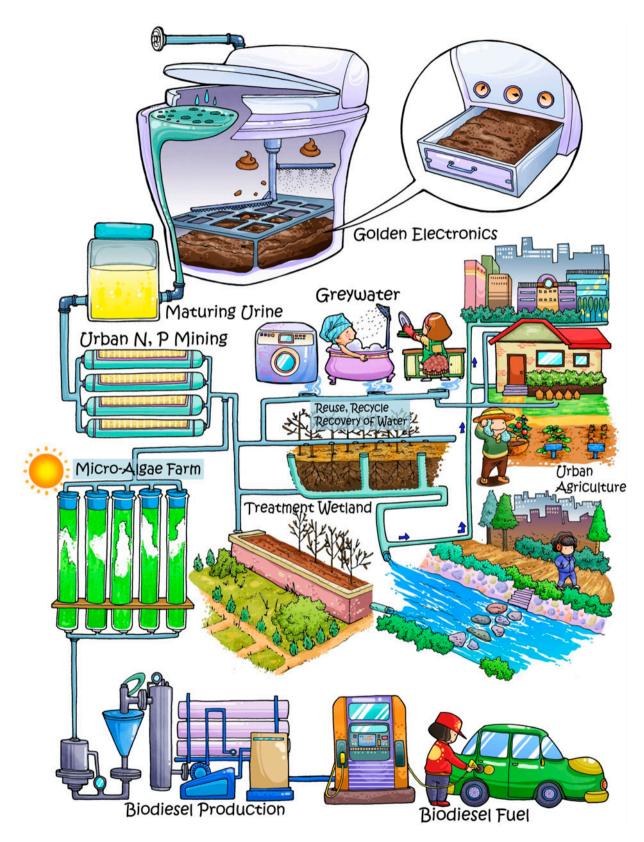


Fig. 6. Schematic representations of various potential engineering concepts merged with existing knowledge and results from the pilot experimental data (titled Science Walden).

6,612 mgN/l), as well as 1/10, 1/100, 1/500, and 1/1,000 diluted solutions. Filtrations were performed using a small flat-sheet unit with an active surface area of  $96.0 \text{ cm}^2$ .

# 3. Results and discussion

#### 3.1. Water quality monitoring

As depicted in Fig. 3, the treatment wetland in the village exhibited very high (~75%), somewhat high, and moderate/low capabilities (~25%) in control of ammonia, DOC, and TN, respectively, through different seasons, even though contaminants levels in the wetland influents somewhat varied and nitrification in the wetlands occurred. There are two reasons why TN removals were relatively low. Firstly, TN levels were relatively low in the influent, thus, the removals of TN, through the constructed wetland, might be also low, and, secondly, ammonium ion included in the influent was believed to be converted to nitrate, thus, the removals of ammonium ion and TN were relatively high and low, respectively. Even with moderate removal efficiencies, the TN in the wetland effluents exhibited levels below a value of 10 mgN/l, which can satisfy both regulation values of TN of small (less than  $50 \text{ m}^3/\text{d}$  capacity) wastewater treatment (with a value of 20 mgN/l) and wastewater recovery (with a value of 10 mgN/l) plants in Korea [21]. Water qualities of the treatment wetland effluents were almost similar to those of adjacent streams (where wetland effluents are supposed to be discharged) which exhibited DOC of 1.2 mgC/l and TN of 0.3 mgN/l. Most of the TN takes the form of nitrates (less toxic than ammonia). In addition, the effluent was recovered to each house to be reused, which may make the concept of zero discharge sanitation and water-quality management system into a reality.

#### 3.2. Community structures in urine and fecal samples

Five major phyla, *Proteobacteria*, *Firmicutes*, *Bacteroidetes*, *Spirochaetes*, and *Actinobacteria*, were identified in the pyrosequenced samples, as shown in Fig. 4. This is in agreement with previous studies presenting that these phyla are dominant with human gut and feces [22–24]. From this analysis, it was confirmed there were no pathogens in the pyrosequenced samples (urine, dried feces, and compost). As hypothesized in the background section focusing on urine, stored urine was identified as a safe nutrients source with respect to pathogens. Furthermore, both dried feces after approximately 1 month storage inside the sanitation toilet with 24 h ventilation as well as compost (mixture of dried feces and garden soil, and composted for approximate for a period longer than 3 months) were also identified as safe to be used with respect to pathogens. *Proteobacteria* were dominant in the urine and dried feces samples, and *Firmicutes* were considered to be originated by soils from gardens [25].

#### 3.3. Salt and nutrient removal efficiencies

As presented in Fig. 5, salts were effectively reduced by the NF membrane filtrations, in terms of conductivity, except with raw urine. Efficiencies of DOC and TN removal were lower than that of conductivity; this implies some possibility of being able to provide nutrients solution, after NF filtration, with certain relatively higher levels of TN and relatively lower levels of conductivity than urine samples before NF filtration. The result of NF filtration depending on the urine dilution rate was shown in Table 1, which presents measured values in terms of conductivity, DOC, and TN.

Certain dilution rate for NF filtration can be selected, depending on cultivating conditions for certain algae species which can be used to produce biofuel.

# 4. Concluding remarks

In this paper, we have combined observed data, experimental pilot test results, and problematic and newly defined water engineering concepts in order to develop an engineering concept or idea which can be practically utilized for urban water management, in conjunction with sanitation, wastewater, and even energy technologies, as schematically depicted in Fig. 6.

# Acknowledgments

This work was supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIP) (No. 2012047029 (TOC) and No. 2011-0030040 (ERC)).

#### References

- [1] Seoul, Statistical Year Book of Seoul, Seoul, 2012.
- [2] S. Hotta, N. Funamizu, Biodegradability of fecal nitrogen in composting process, Bioresour. Technol. 98 (2007) 3412–3414.
- [3] T. Karak, P. Bhattacharyya, Human urine as a source of alternative natural fertilizer in agriculture: A flight of fancy or an achievable reality, Resour. Conserv. Recy. 55 (2011) 400–408.

- [4] C. Polprasert, Organic Waste Recycling, Wiley, Chichester, 1996.
- [5] N.L. Schouw, S. Danteravanich, H. Mosbæk, J.C. Tjell, Composition of human excreta—a case study from Southern Thailand, Sci. Total. Environ. 286 (2002) 155– 166.
- [6] B. Vinnerås, A. Nordin, C. Niwagaba, K. Nyberg, Inactivation of bacteria and viruses in human urine depending on temperature and dilution rate, Water. Res. 42 (2008) 4067–4074.
- [7] K.M. Udert, T.A. Larsen, M. Biebow, W. Gujer, Urea hydrolysis and precipitation dynamics in a urine-collecting system, Water Res. 37 (2003) 2571–2582.
- [8] IWA, Constructed Wetlands for Pollution Control. Processes, Perfomance, Design and Operation, IWA publishing, London, 2000.
- [9] A. Gross, O. Shmueli, Z. Ronen, E. Raveh, Recycled vertical flow constructed wetland (RVFCW)—a novel method of recycling greywater for irrigation in small communities and households, Chemosphere 66 (2007) 916–923.
- [10] Z. Li, H. Gulyans, M. Jahn, D.R. Gajurel, R. Otterpohl, Greywater treatment by constructed wetlands in combination with TiO<sub>2</sub>-based photocatalytic oxidation for suburban and rural areas without sewer system, Water. Sci. Technol. 48 (2004) 101–106.
- [11] P.A. Vesilind, A.E. Rimer, Unit Operations in Resource Recovery Engineering, Prentice-Hall, Englewood Cliffs, NJ, 1981.
- [12] J. Vymazal, The use constructed wetlands with horizontal sub-surface flow for various types of wastewater, Ecol. Eng. 35 (2009) 1–17.
- [13] S. Cho, T.T. Luong, D. Lee, Y.-K. Oh, T. Lee, Reuse of effluent water from a municipal wastewater treatment plant in microalgae cultivation for biofuel production, Bioresour. Technol. 102 (2011) 8639–8645.
- [14] A. Ludwig, Create an Oasis with Greywater, Oasis Design, Santa Barbara, CA, 2007.
- [15] A.H. Franks, H.J. Harmsen, G.C. Raangs, G.J. Jansen, F. Schut, G.W. Welling, Variations of bacterial populations in human feces measured by fluorescent *in situ* hybridization with group-specific 16S rRNA-targeted

oligonucleotide probes, Appl. Environ. Microbiol. 64 (1998) 3336–3345.

- [16] H. Hayashi, M. Sakamoto, M. Kitahara, Y. Benno, Molecular analysis of fecal microbiota in elderly individuals using 16S rDNA library and T-RFLP, Microbiol. Immunol. 47 (2002) 557–570.
- [17] S.K. Pradhan, A.M. Nerg, A. Sjöblom, J.K. Holopainen, H. Heinonen-Tanski, Use of human urine fertilizer in cultivation of cabbage (*Brassica oleracea*)—impacts on chemical, microbial, and flavor quality, J. Agric. Food. Chem. 55 (2007) 8657–8663.
- [18] E.G. Zoetendal, A.D. Akkermans, W.M. De Vos, Temperature gradient gel electrophoresis analysis of 16S rRNA from human fecal samples reveals stable and host-specific communities of active bacteria, Appl. Environ. Microbiol. 64 (1998) 3854–3859.
- [19] S. Lee, G. Park, G. Amy, S.K. Hong, S.-H. Moon, D.H. Lee, J. Cho, Determination of membrane pore size distribution using the fractional rejection of nonionic and charged macromolecules, J. Membr. Sci. 201 (2002) 191–201.
- [20] Y. Shim, H.J. Lee, S. Lee, S.H. Moon, J. Cho, Effects of natural organic matter and ionic species on membrane surface charge, Environ. Sci. Technol. 36 (2002) 3864– 3871.
- [21] Korean Ministry of Environment, Water Quality and Ecosystem Conservation Act, Korean Government, Seoul, 2010.
- [22] P.B. Eckburg, E.M. Bik, C.N. Bernstein, E. Purdom, L. Dethlefsen, M. Sargent, S.R. Gill, K.E. Nelson, D.A. Relman, Diversity of the human intestinal microbial flora, Science 308 (2005) 1635–1638.
- [23] S.R. Gill, M. Pop, R.T. DeBoy, P.B. Eckburg, P.J. Turnbaugh, B.S. Samuel, J.I. Gordon, D.A. Relman, C.M. Fraser-Liggett, K.E. Nelson, Metagenomic analysis of the human distal gut microbiome, Science 312 (2006) 1355–1359.
- [24] Y.D. Nam, M.J. Jung, S.W. Roh, M.S. Kim, J.W. Bae, Comparative analysis of Korean human gut microbiota by barcoded pyrosequencing, PLoS One 6 (2011) e22109.
- [25] L. Zhang, Z. Xu, Assessing bacterial diversity in soil, J. Soils. Sed. 8 (2008) 379–388.