



23 August 2019

Symposium on Hydro-microbiological Approach for Water Security in Kathmandu Valley, Nepal



Outcomes of JICA WaSH-Mia/SATREPS Project



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Symposium on Hydro-microbiological approach for water security in Kathmandu Valley, Nepal Outcomes of JICA WaSH-Mia/SATREPS Project

Date: 23 August 2019, Venue: Hotel Himalaya, Kupondole, Lalitpur

Tentative program:

9:15-10:00 Registration

10:00 – 11:00 Opening Session chaired by Joint Secretary, MoWS

- Welcome speech and introduction of the project by Project Managers
- Lighting Lamp Ceremony by the chief guest, Ambassador of Japan
- Opening remarks
 - Dean, IOE, TU
 - Secretary, NAST
- Remark by the chief guest, Ambassador of Japan
- Remark by the opening session chair, Joint Secretary, MoWS

11:00-13:20 Morning presentation session, chaired by Mr. Pramod S. S. Pradhan, AITM

- Water situation of Kathmandu Valley
 - Water quantity by WG1 (40min)
 - Water quality by WG2 (30min)
 - Microbial pollution by WG3 (30min)
 - Water use for daily life by WG5 (25min)
 - Q & A (15min)

13:20-14:20 Lunch in the hotel dining hall

14:20-16:05 Afternoon presentation session, chaired by Dr. Kei Nishida, UY

- Locally fitted, Compact and Distributed (LCD) water treatment system
 - Structure and performance by WG4 (30min)
 - User's evaluation of LCD by WG5 (15min)
 - Q & A (15min)
- Water future in Kathmandu
 - Utilization of water security map by Task Force (10min)
 - Strategy for water security in Kathmandu by Task Force/KVWSMB (20min)
 - Q & A (15min)

16:05-16:20 Closing

- Closing remark by JICA Chief Representative
- Closing address by Project Managers

16:20- 17:00 Afternoon Coffee/tea (the end of the symposium)

Project Summary

Scientists have been urged to tackle global issues - Energy and Water, Climate Change and Sustainability. To address water security issues in developing countries where limited energy and water resources were available, "The project for Hydro-microbiological approach for water security in Kathmandu Valley, Nepal" was formed in May 2014 as a Japan-Nepal cooperation project under SATREPS¹ framework of JICA/JST. From Nepalese side, Ministry of Water Supply, IOE/IOM/CDG of Tribhuvan Uni., KWWSMB, KUKL, AITM, DHM, NAST, Crew and SEN participated in this project, while Uni. of Yamanashi took a leading role in Japanese side.

This project was implemented by five Working Groups (WG) for the purposes to "create a water security map by applying water safety diagnostic technology" and to "maximize local water purification potential of the Kathmandu Valley". For the application of water diagnostic technology, activities of 'water resource diagnosis', 'water quality diagnosis', 'Microbial / Public Health Diagnosis' and 'Social impact assessment' were conducted by WG1, WG2, WG3 and WG5 respectively. WG4 carried out research and development of water treatment technologies such as sponge sloping water treatment, sand filtration water treatment, biofilm water treatment and constructed wetland water treatment.

More specifically, WG1 studied potable water resources situation, including water demand, consumption and supply in Kathmandu Valley, forecasted future situation and elaborated reports on 'spatio-temporal distribution of water resources and the long-term variation' and 'possibilities of developing alternative techniques to utilize water resources'. Regarding water quality diagnosis, the situation and sources of groundwater pollution were studied by WG2, while WG3 studied Microbiological situation of environmental water, such as groundwater, surface water, and rainwater in the Kathmandu Valley. Under the research of water treatment technology by WG4, a prototype locally fitted, compact and distributed (LCD) water treatment system for community use in line with the local situation was developed. The targeted qualities of the treated water were the levels of drinking use of iron, ammonium nitrogen, nitrate nitrogen, and for domestic use of turbidity as regulated in Nepal Drinking Water Quality Guideline Standard. WG5 was responsible for social impact assessment and social and economic evaluation for the purpose of implementation and installation of the LCD water treatment system. Based on the findings of WG1,2,3 and 5, the integrated water security maps of potable water resources, water quality, water borne infections, and water stress were elaborated.

Besides the above, a task force was organized to enhance the social implementation of the project outcomes, particularly for the integrated water security map and the LCD technology. Capacity building of researchers and technology transfer to stakeholders by the research activities, the on-site collaboration and training programs in Japan were also important part of the project, through which eight Nepalese students got PhD degree.

It is hoped that the project outcomes would be benefit to academia, government / private water sectors and people in Kathmandu Valley, and in the future, other localities and countries whose water situation is similar to Kathmandu's.

¹ Science and Technology Research Partnership for Sustainable Development

Report of JICA WaSH-Mia/SATREPS Project

for Symposium on Hydro-microbiological approach
for water security in Kathmandu Valley, Nepal



23 August 2019



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I.) Project Introduction

Background and purpose

The project 'Hydro-microbiological approach for water security in the Kathmandu Valley' was approved for funding under the SATREPS program in 2013. SATREPS is a Japanese government program that promotes international joint research targeting global issues and are expected to lead to outcomes with potential for practical utilization, and to enhance research capacity in the recipient country. For this project, the University of Yamanashi in Japan partnered with Institute of Engineering of Tribhuvan University and other government and non-government institutes.

The project targeted the development of the technique to ensure water security in the Kathmandu Valley, Nepal by integrating microbiology, hydrology and water chemistry. The main purpose of the project was to ensure 'water security' from human health perspective by enhancing the management system of the potable water resources such as shallow and deep groundwater, surface and rainwater. At the same time, it aimed to develop a locally-fitted compact and distributed (LCD) water treatment system by fully utilizing microbial potential onsite that can be applicable to other areas as well.

Working groups

The project had an integrated framework of 'hydro-microbiology' and consists of five 'working groups':

1. WG1: Water resource assessment

(Aim: To study and forecast potable water resources situation, including water demand, consumption and supply in the Kathmandu Valley)

2. WG2: Water quality assessment

(Aim: To study water pollution situation of shallow and deep groundwater and surface and rain water)

3. WG3: Microbial and public health assessment

(Aim: To study microbiological situation of shallow and deep groundwater, and surface rain water in the Kathmandu Valley)

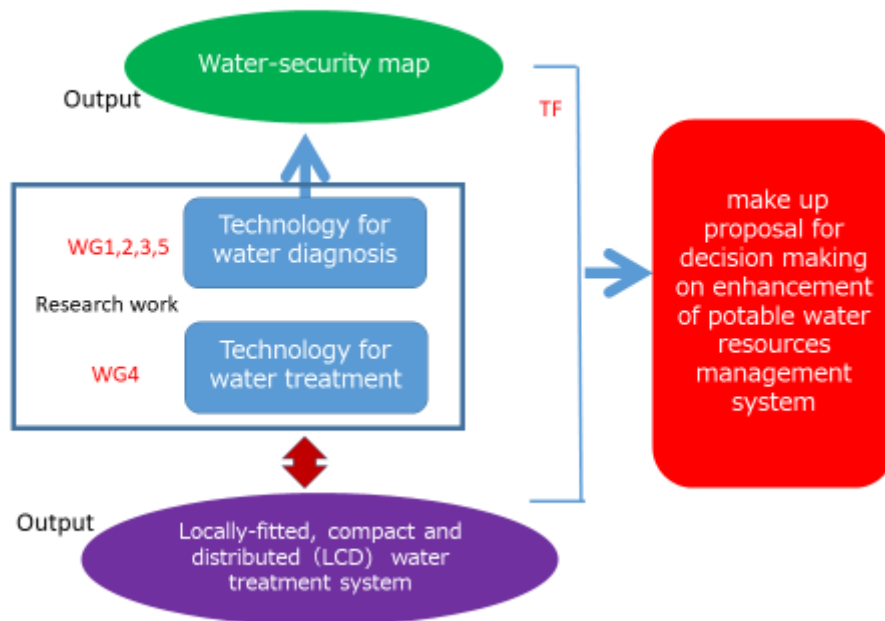
4. WG4: Water treatment system development

(Aim: To develop appropriate LCD treatment system for groundwater and surface water in the Kathmandu Valley)

5. WG5: Socio-economic assessment

(Aim: To study social and economic evaluation of LCD treatment system installation in the Kathmandu Valley)

Integrated framework among the working groups (WGs)



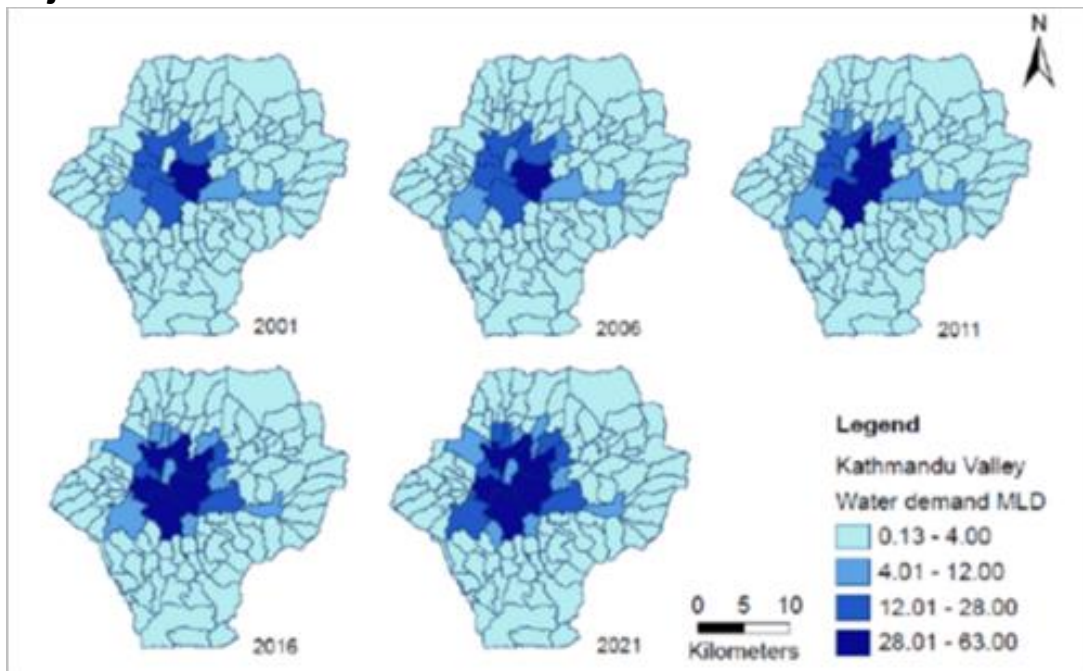
Main members of SATREPS project

Nepalese side	Japanese side
Project Managers	
Dr. Narendra Man Shakya	Dr. Futaba Kazama
Group Leaders	
Working Group 1	
Dr. Narendra Man Shakya	Dr. Hiroshi Ishidaira
Working Group 2	
Dr. Suresh Das Shrestha	Dr. Takashi Nakamura
Working Group 3	
Dr. Jeevan B. Sherchand	Dr. Eiji Haramoto
Working Group 4	
Dr. Iswar Man Amatya	Dr. Tadashi Tohyama
Working Group 5	
Mr. Chok Dhital	Dr. Junko Shindo

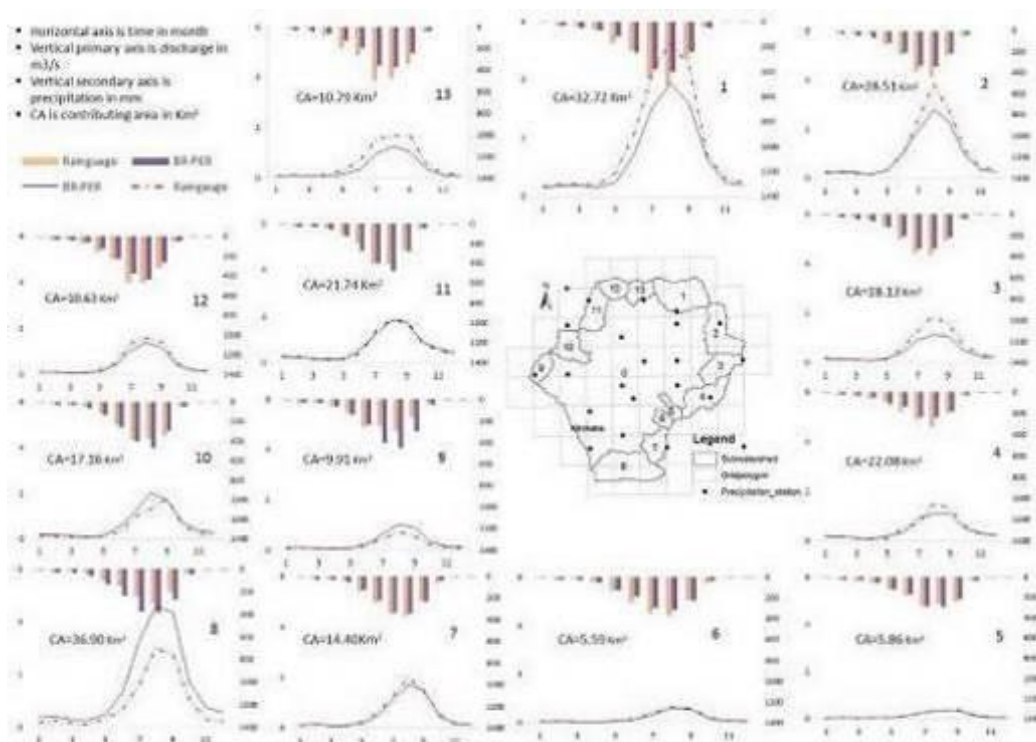
II.) Achievements of working groups (WGs)

Working Group 1 (Water resource assessment)

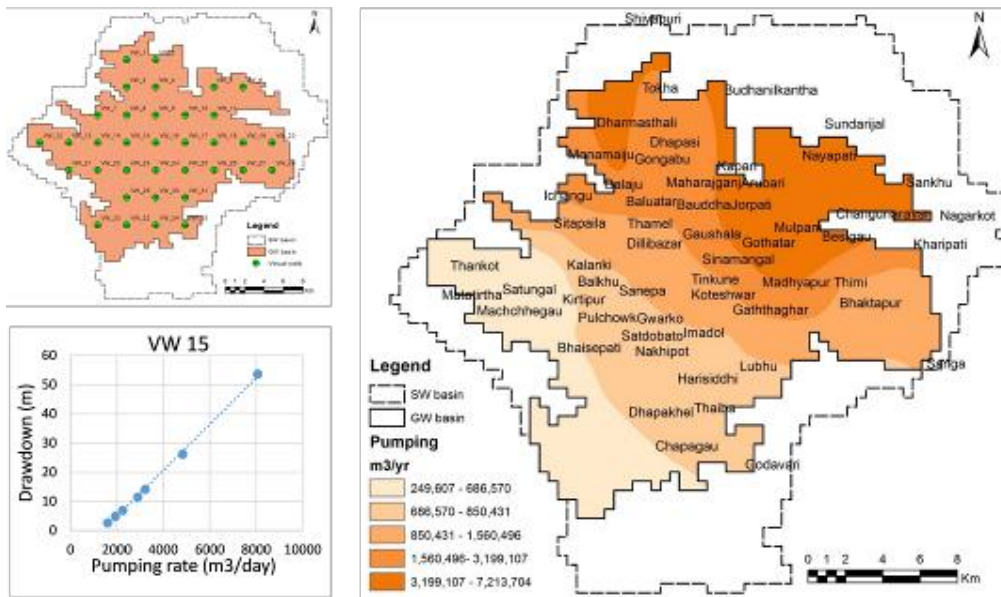
A.) Major results



a. Water demand distribution by municipalities and village development committees

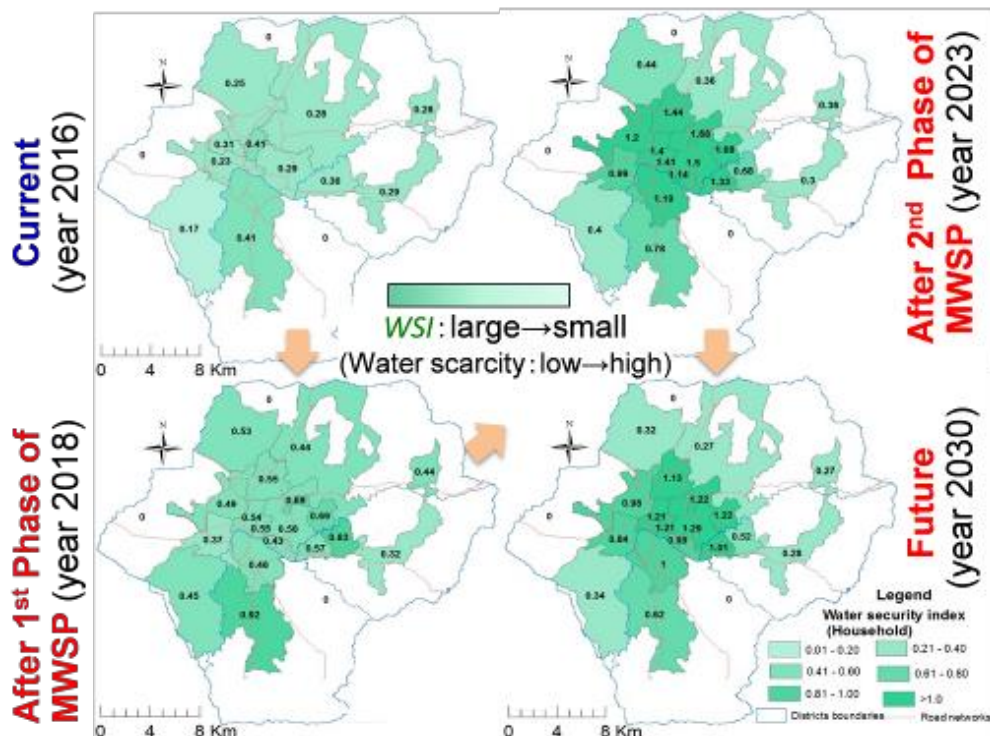


b. Estimation of river flow from mountain areas using distributed runoff model



c. Estimation of deep groundwater abstraction potential

{Upper left: Arrangement of virtual pumping wells; Lower left: corresponding to the scenario (amount of pumping) calculation example of change of groundwater fall amount; Right: Spatial distribution of deep groundwater pumping capacity}



d. Spatio-temporal distribution of water security index [ratio of KUKL water supply (S) to demand (D): WSI]

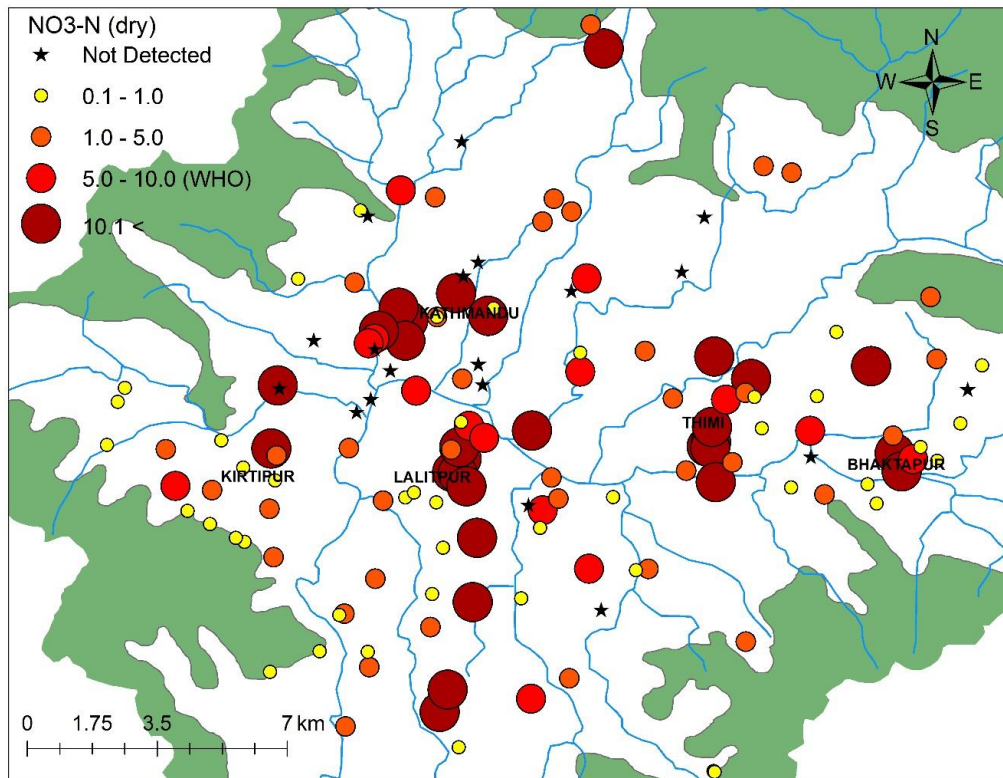
B.) Status of technology transfer to the counter part

In 2014, one JICA short-term trainee (Master's student at Tribhuwan University) was accepted to provide technical guidance on trend analysis of hydrological data, groundwater model construction, land use data analysis and GIS use. In 2015, one short-term JICA trainee (1 student at IOE, Tribhuwan University) was accepted to provide technical guidance on trend analysis of hydrological meteorological data, groundwater model construction, land use data analysis, and GIS use.

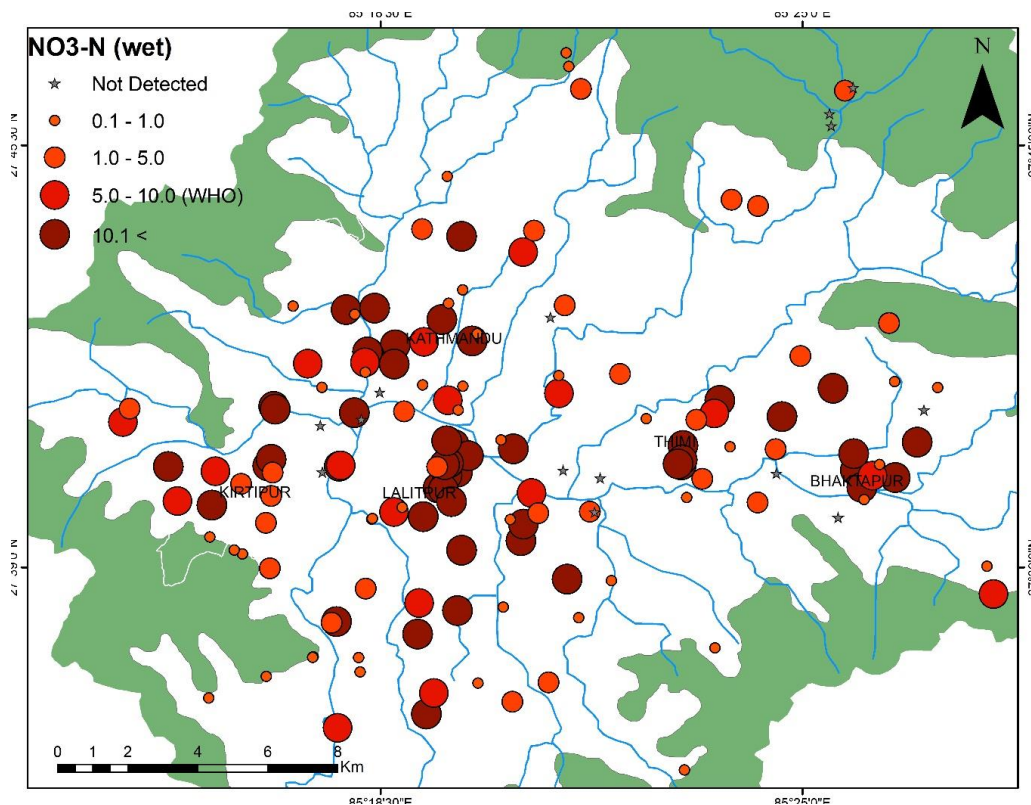
In 2016, we accepted two JICA short-term trainees (two students from IOE, Tribhuwan University) and provided technical guidance on trend analysis of hydrological data, groundwater model construction, land use data analysis, and GIS use. In 2018, one short-term JICA trainee (Tribhuwan University lecturer) was accepted to provide technical guidance on constructing a groundwater model, analyzing land use data, and using GIS.

Working group 2 (Water quality assessment)

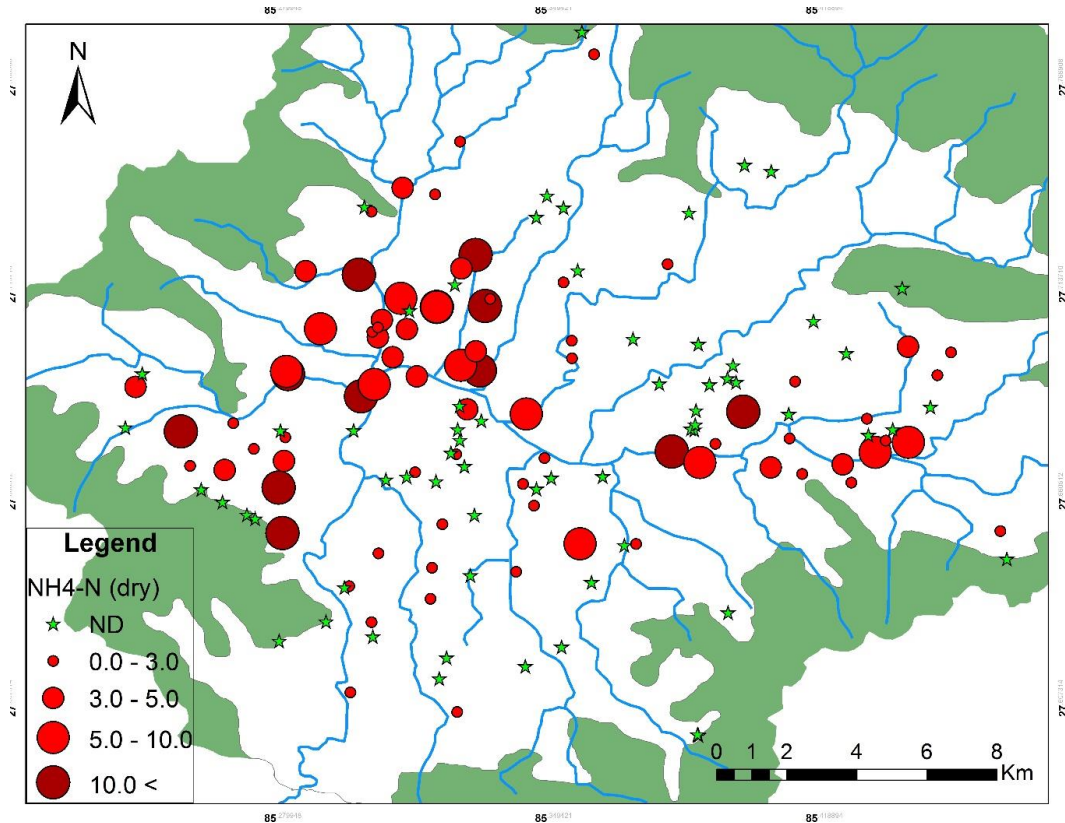
A.) Major results



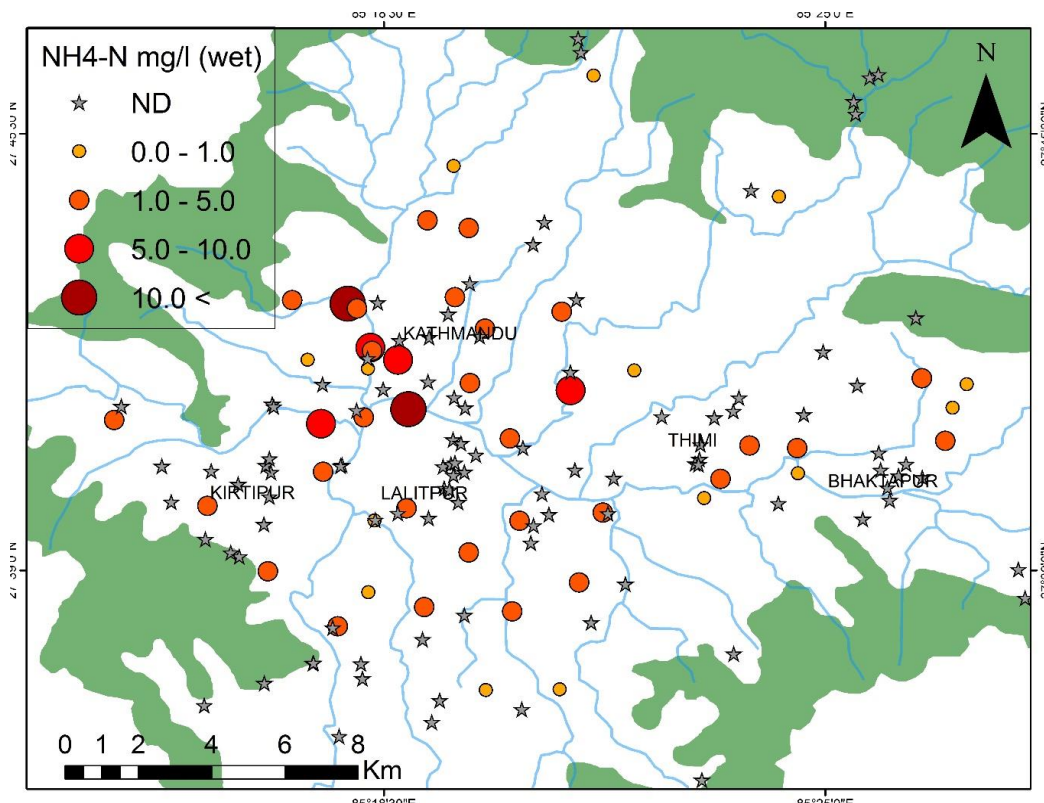
a) Nitrate-nitrogen concentration (mg/L) in shallow groundwater of 2016 dry season.



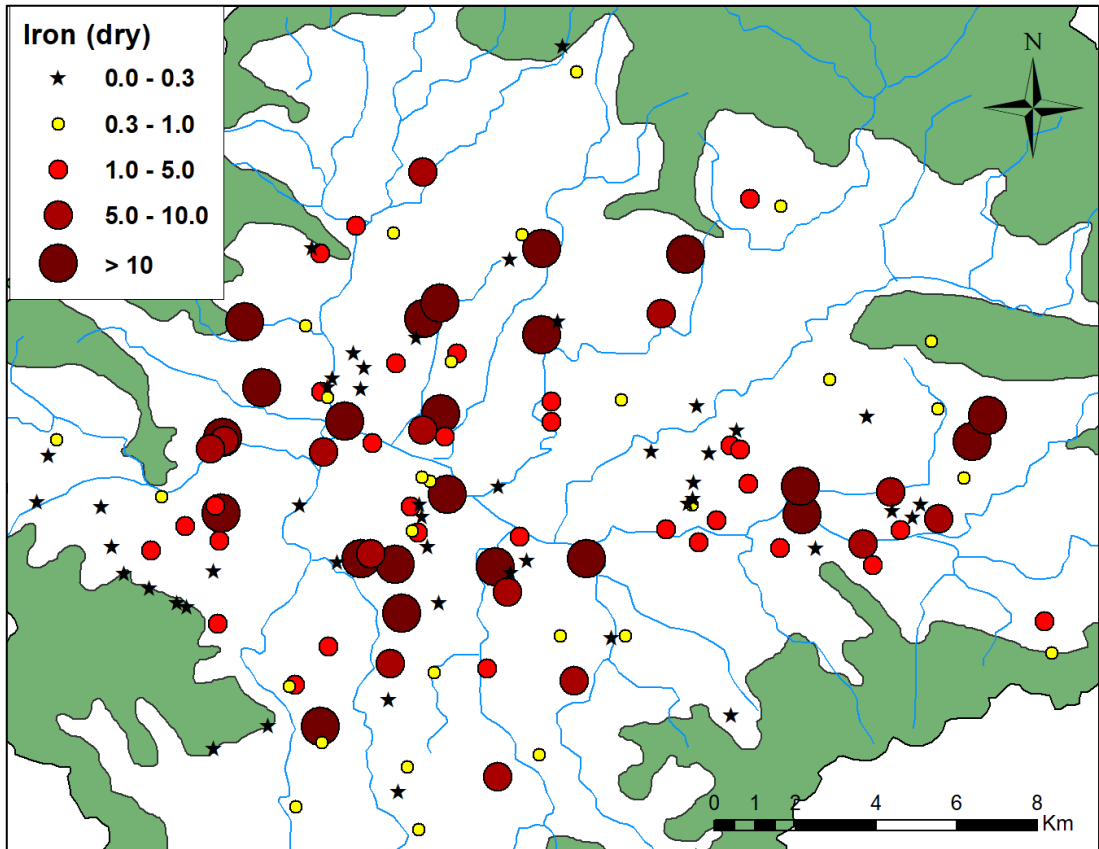
b) Nitrate-nitrogen concentration (mg/L) in shallow groundwater of 2016 wet season.



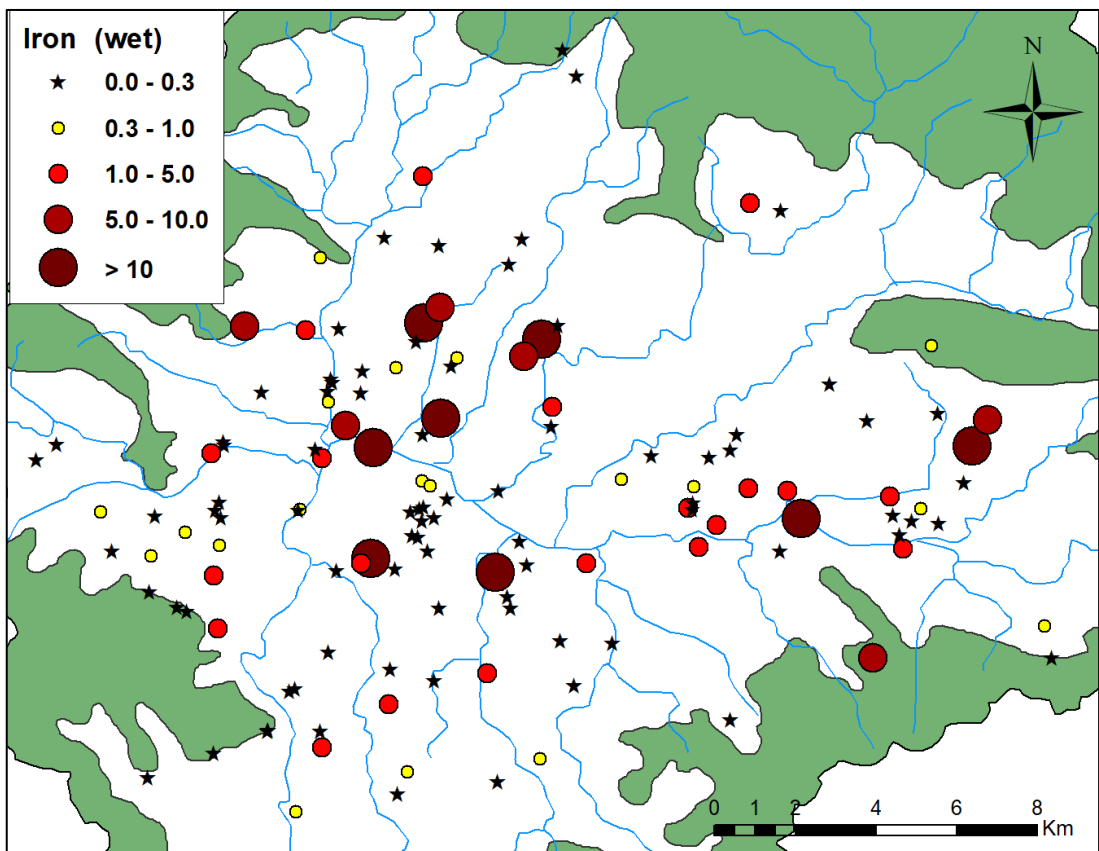
c) Ammonium-nitrogen concentration (mg/L) in shallow groundwater of 2016 dry season.



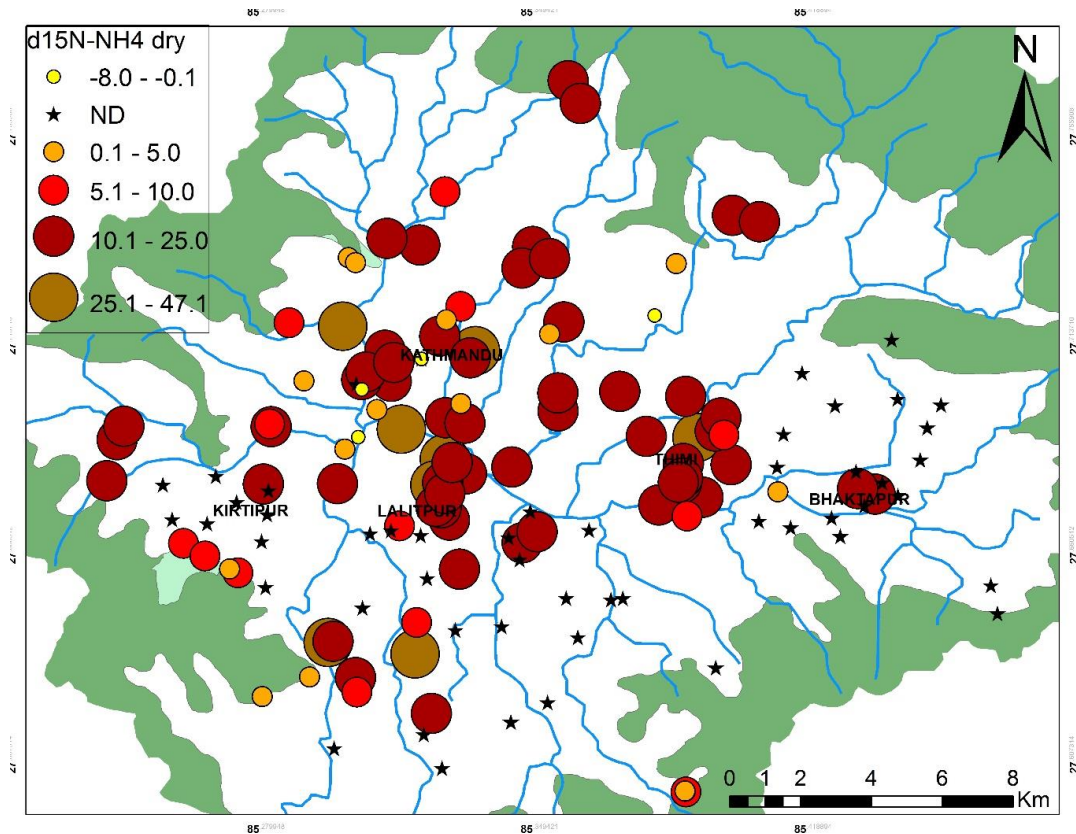
d) Ammonium-nitrogen concentration (mg/L) in shallow groundwater of 2016 wet season.



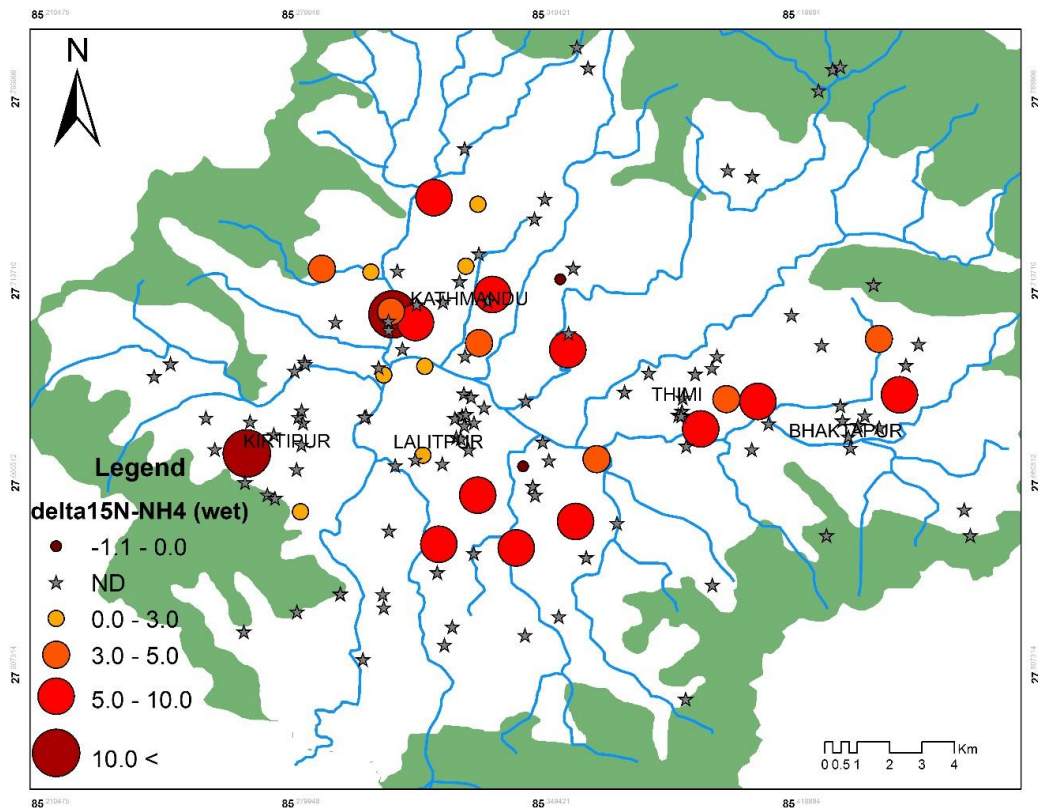
e) Iron concentration (mg/L) in shallow groundwater of 2016 dry season.



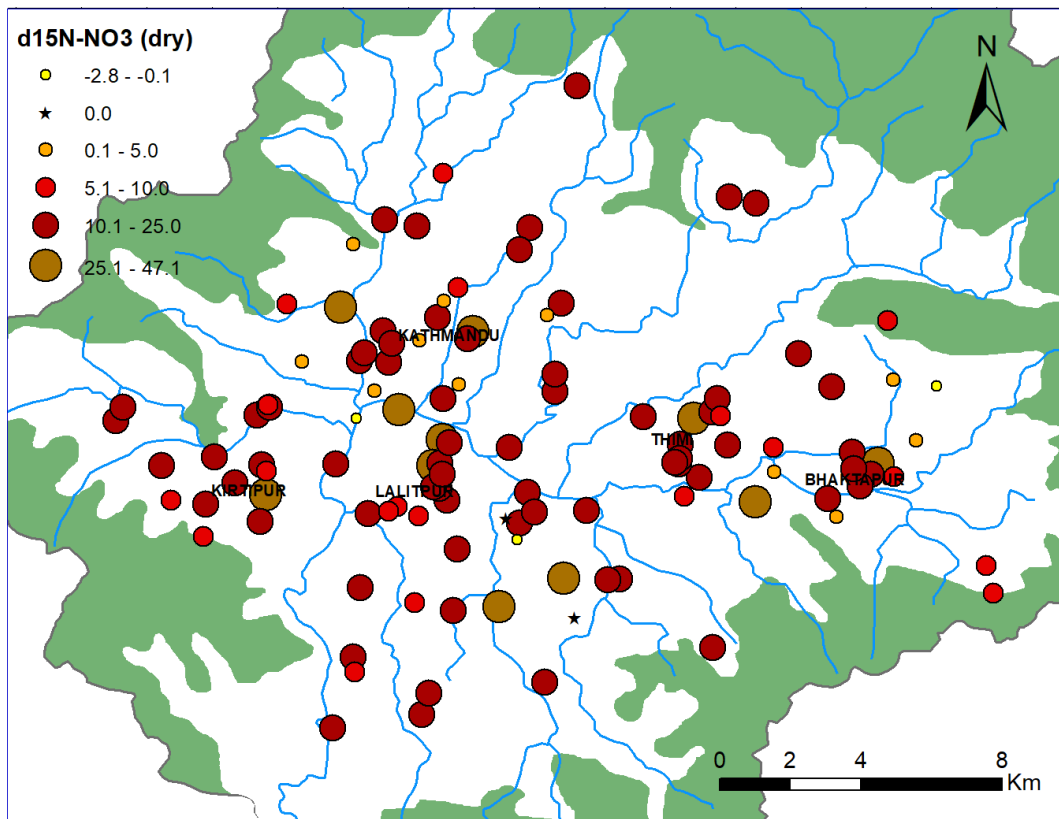
f) Iron concentration (mg/L) in shallow groundwater of 2016 wet season.



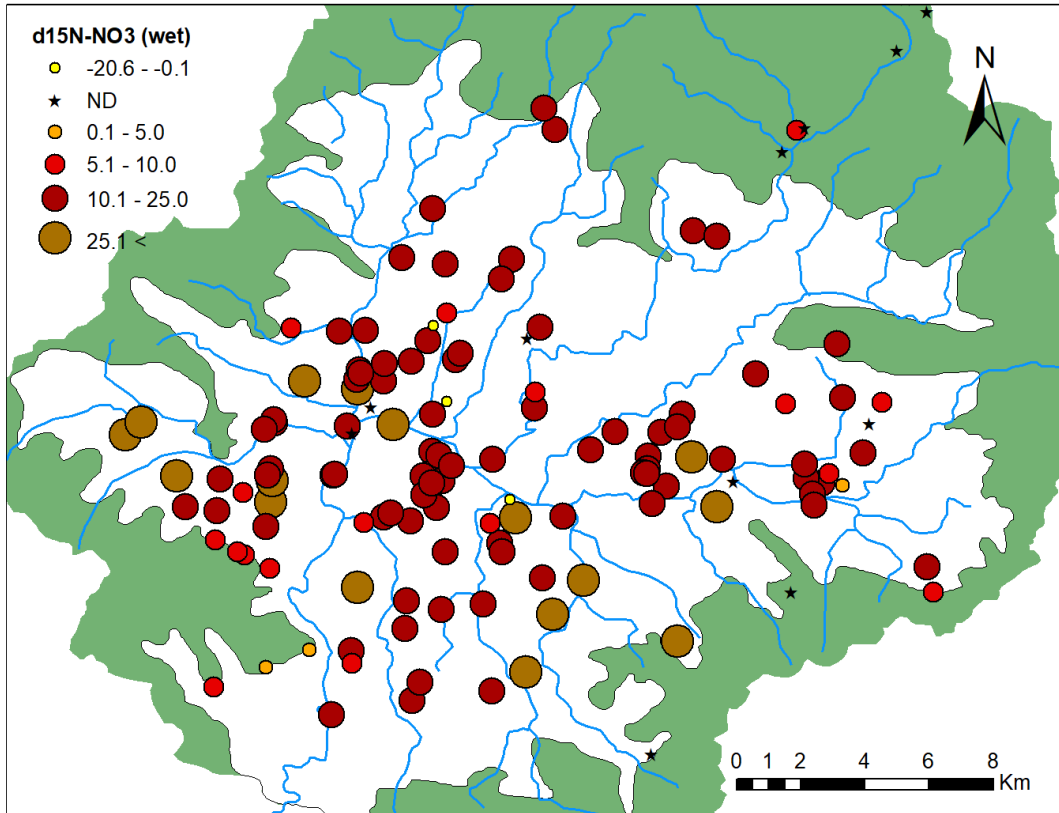
g) Ammonium-nitrogen stable isotope values (‰) in shallow groundwater of 2016 dry season.



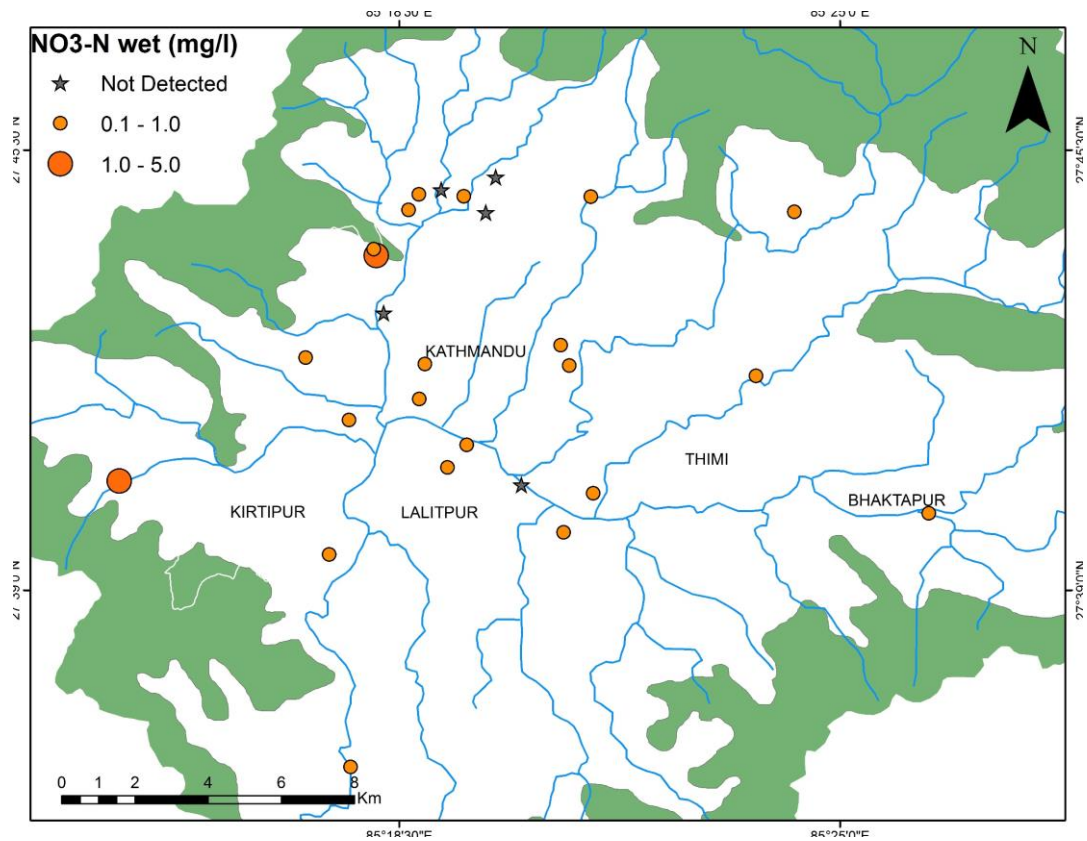
h) Ammonium-nitrogen stable isotope values (‰) in shallow groundwater of 2016 wet season.



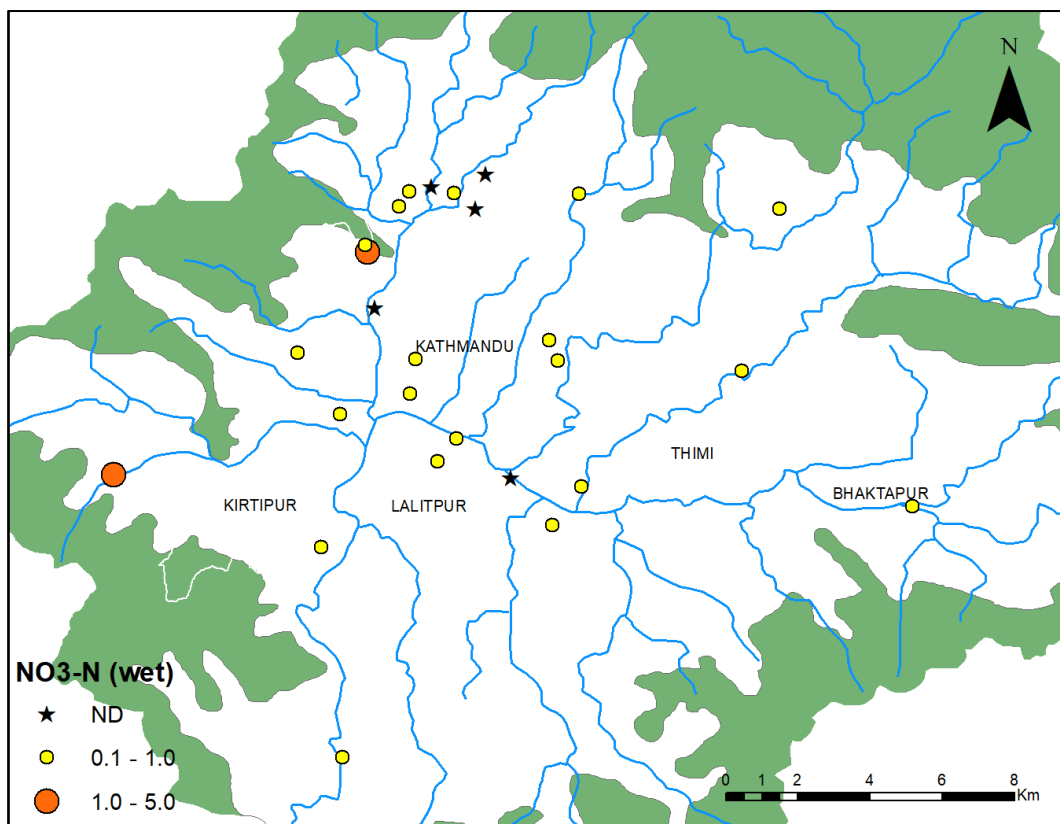
i) Nitrate-nitrogen stable isotope values (‰) in shallow groundwater of 2016 dry season.



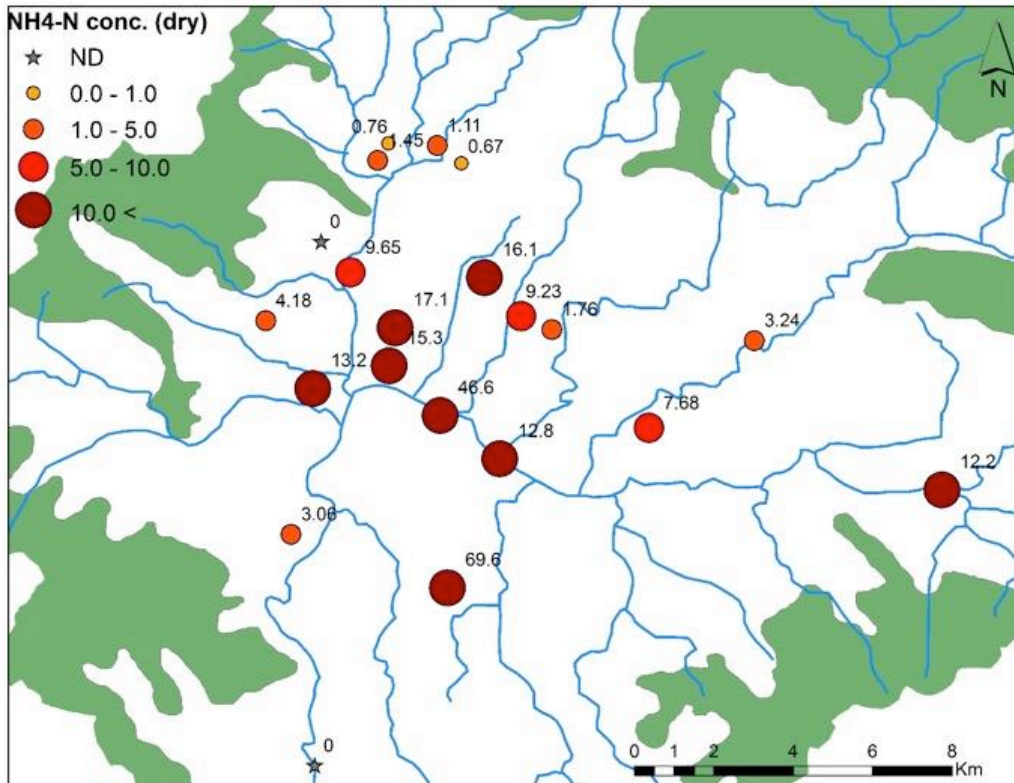
j) Nitrate-nitrogen stable isotope values (‰) in shallow groundwater of 2016 wet season.



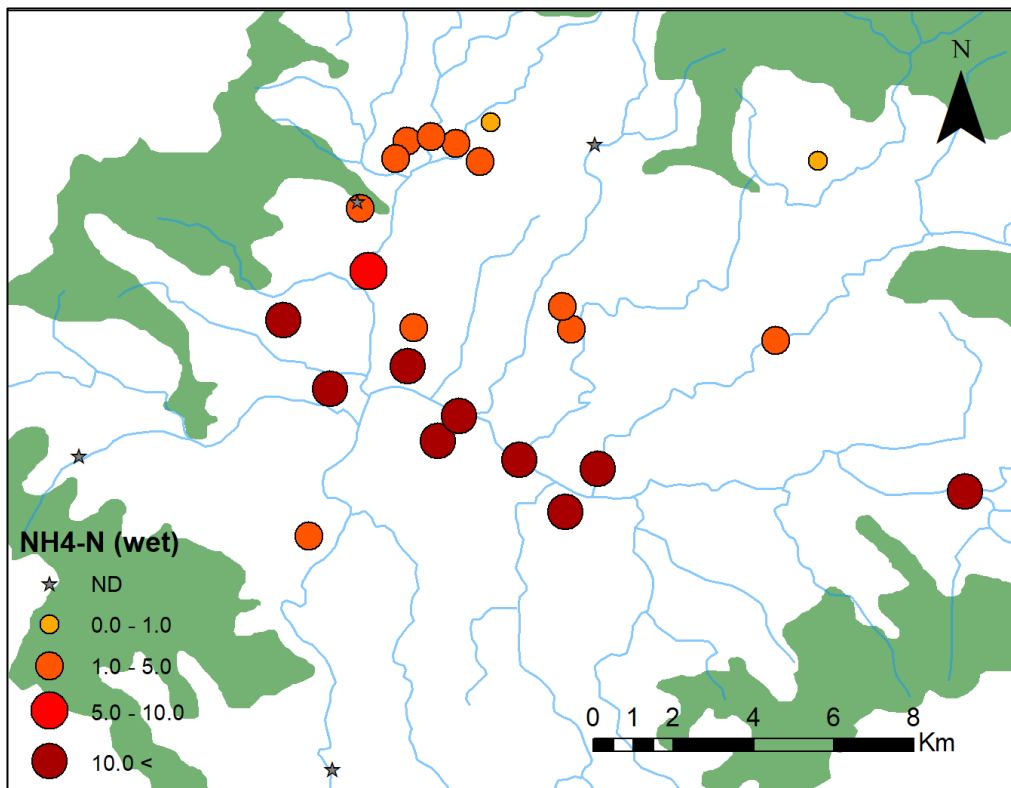
k) Nitrate-nitrogen concentration (mg/L) in Deep groundwater of 2016 dry season.



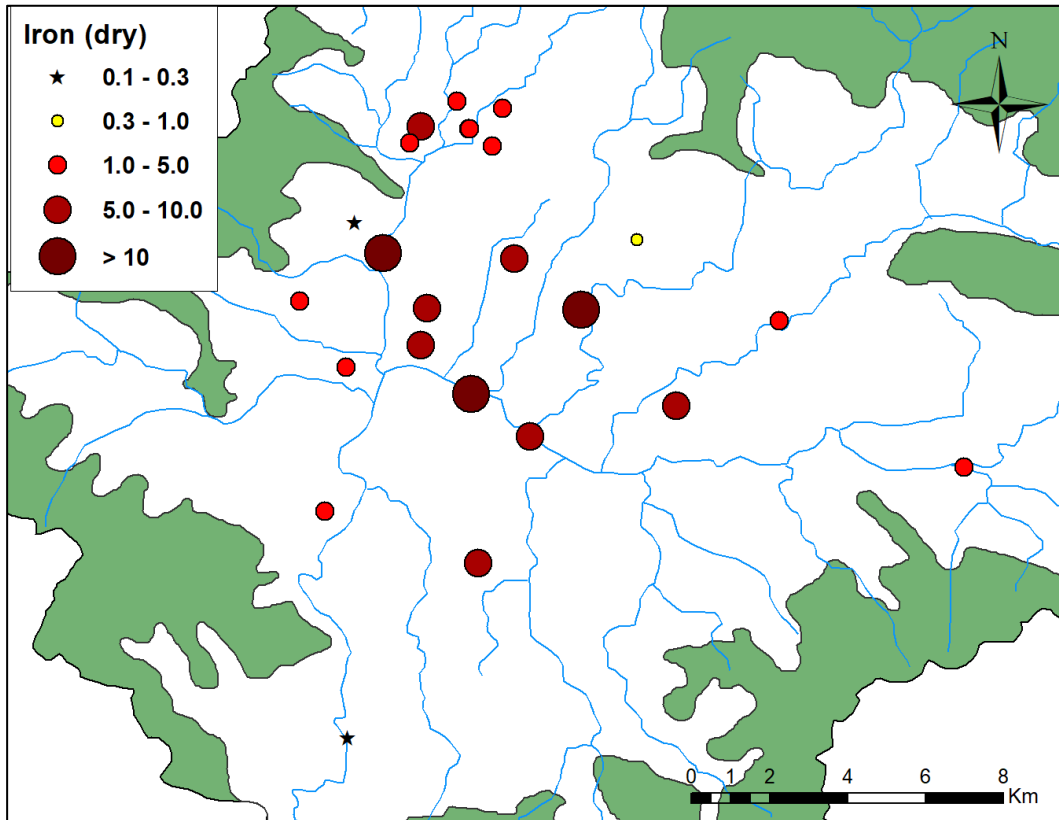
l) Nitrate-nitrogen concentration (mg/L) in Deep groundwater of 2016 wet season.



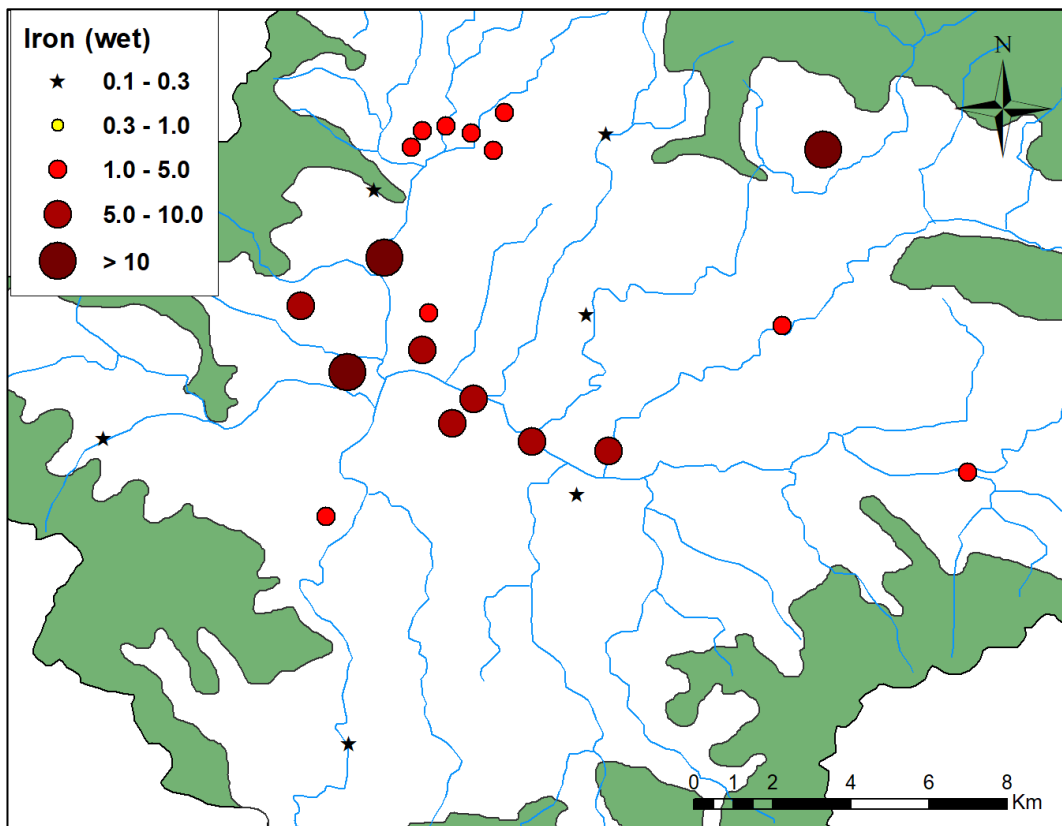
m) Ammonium-nitrogen concentration (mg/L) in Deep groundwater of 2016 dry season.



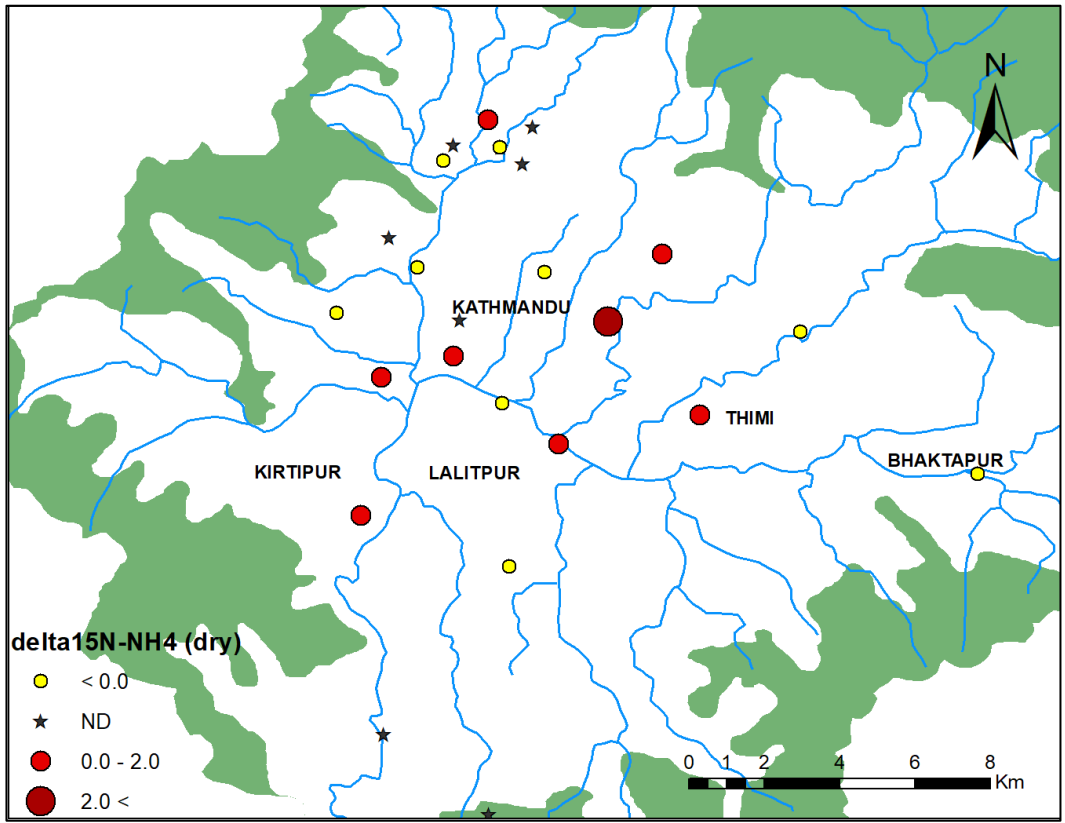
n) Ammonium -nitrogen concentration (mg/L) in Deep groundwater of 2016 wet season.



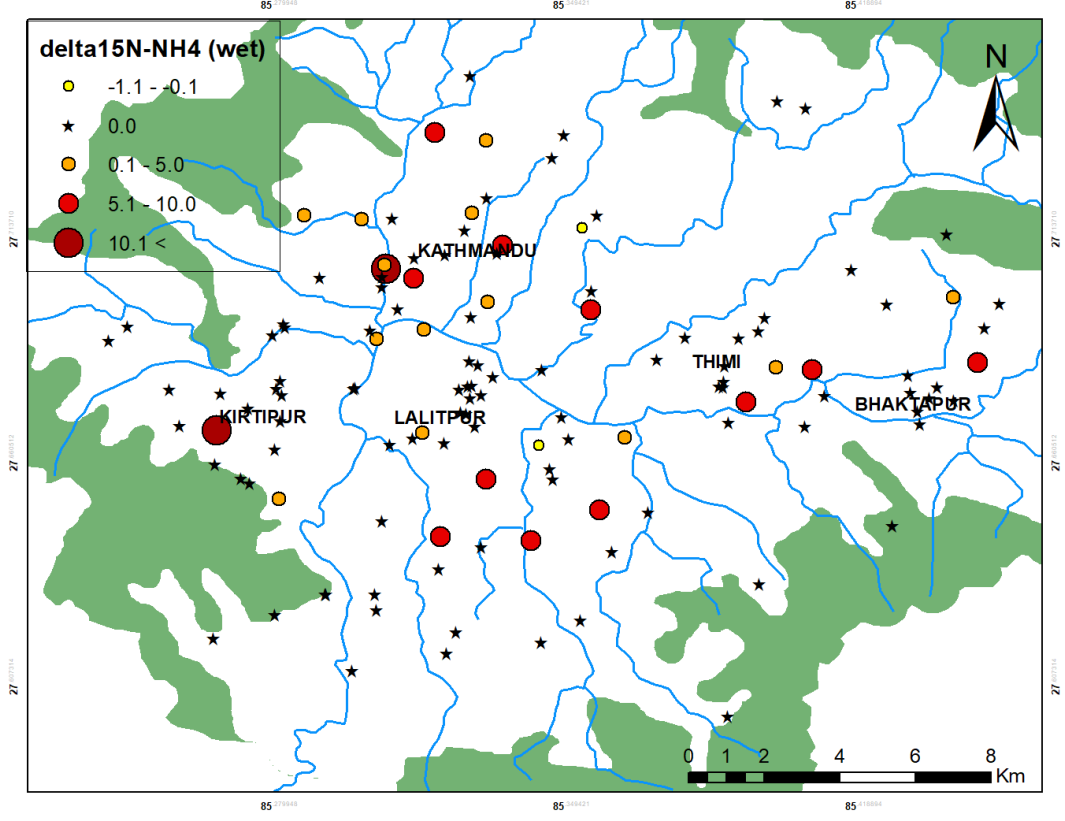
o) Iron concentration (mg/L) in Deep groundwater of 2016 wet season.



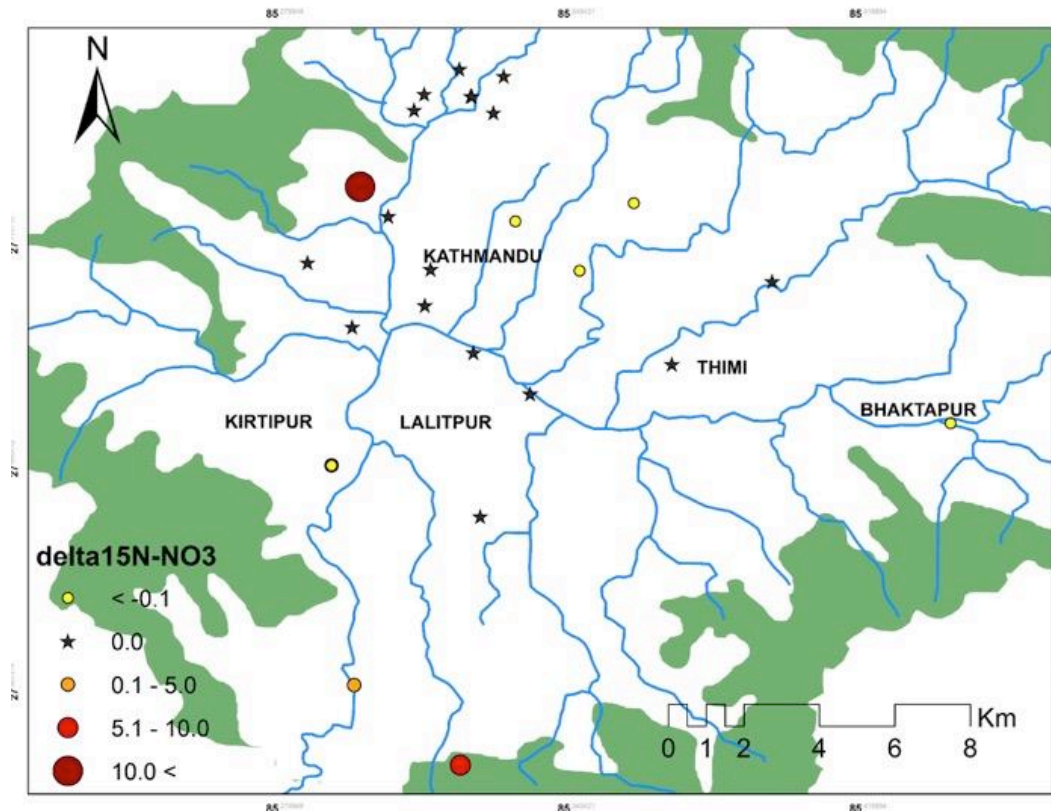
p) Iron concentration (mg/L) in Deep groundwater of 2016 wet season.



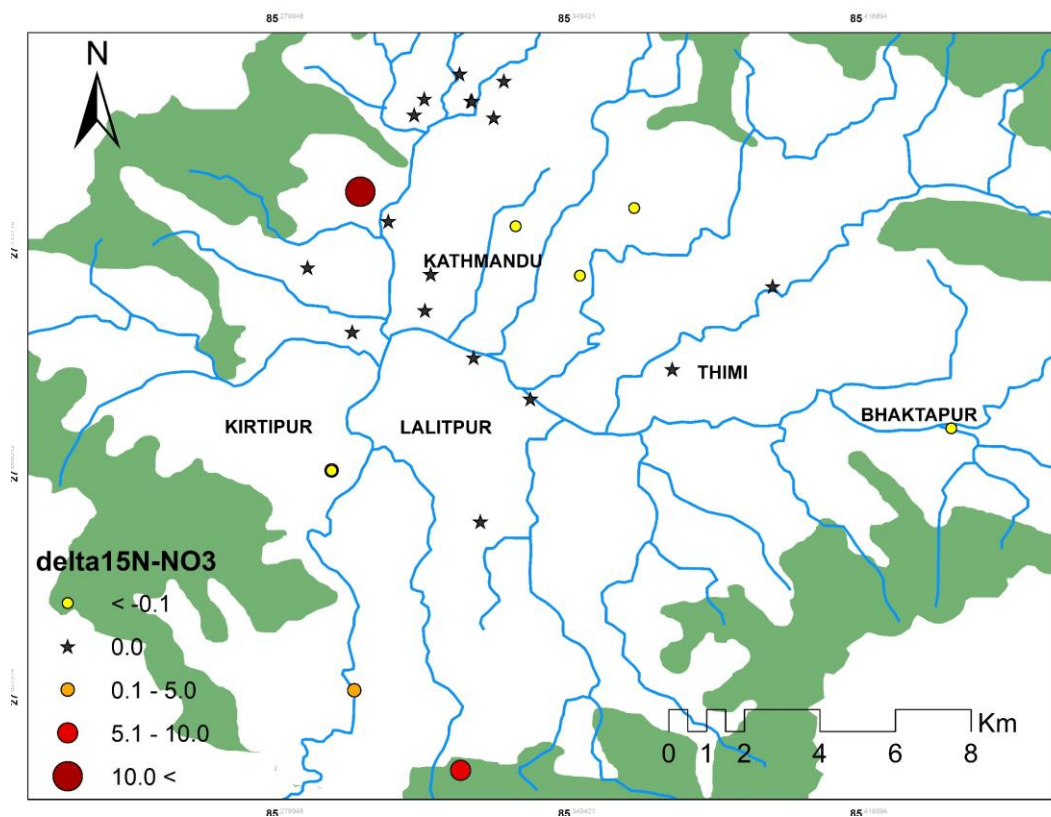
q) Ammonium-nitrogen stable isotope values (‰) in deep groundwater of 2016 dry season.



r) Ammonium-nitrogen stable isotope values (‰) in deep groundwater of 2016 wet season.



s) Nitrate-nitrogen stable isotope values (‰) in deep groundwater of 2016 dry season.



t) Nitrate-nitrogen stable isotope values (‰) in deep groundwater of 2016 wet season.

B.) Status of technology transfer to the counter parts

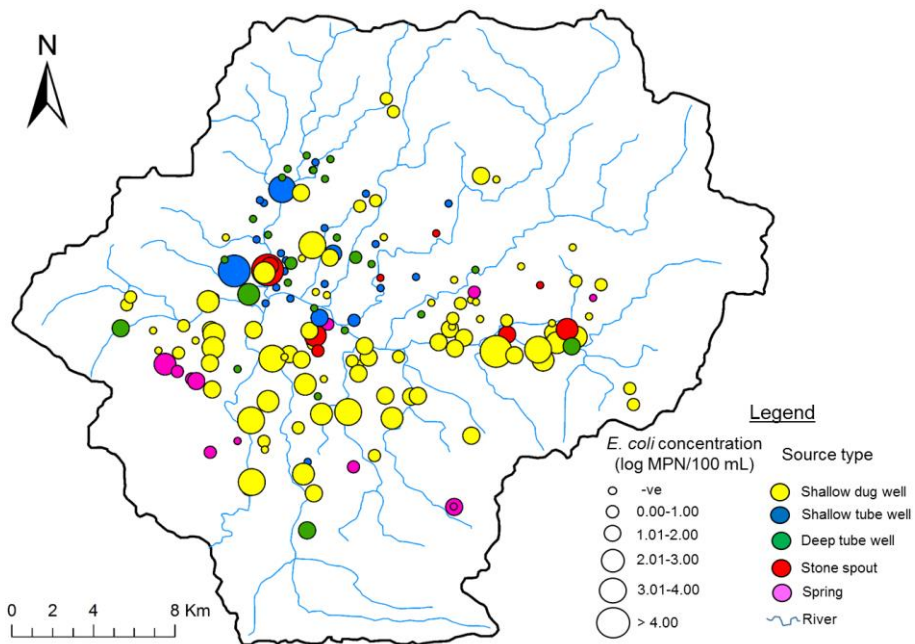
In order to conduct a survey covering the entire area of the Kathmandu Valley, the formation of a field survey team by the students and staffs of the counterparts was essential. Therefore, continuous technical training and technology transfer for on-site surveys were conducted for 24 students of the counterpart (in Tribhuvan University, Faculty of Geology) in 2014, and 50 students in 2015.

By carrying out technology transfer, we shared on-site survey methods such as setting of water sampling point, sampling of samples, local water quality analysis, measurement of groundwater level, and interview survey of water use situation. Furthermore, the details of management of these field survey methods and collected samples and data were prepared in a manual, and a system was established in which multiple teams can conduct surveys simultaneously. In the survey of the whole basin carried out in the dry season of 2015, the selection of the survey well, the series of the survey plan and the field survey were conducted with the initiative of the counterpart, and the regional survey was completed. Furthermore, apart from the full-scale survey, sustainability is also being secured, and technology transfer such as field survey and local water quality analysis is almost completed.

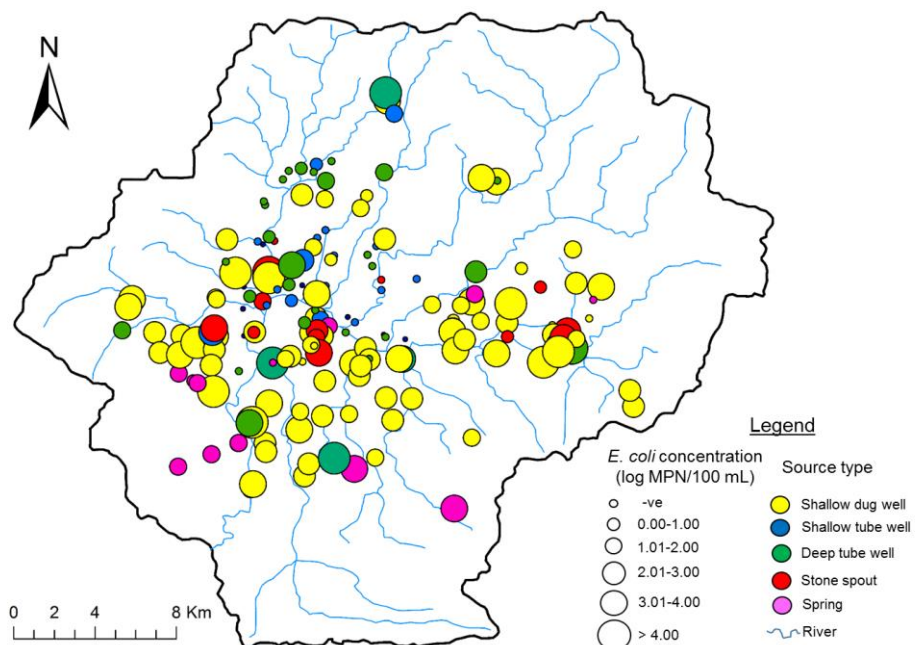
In 2016, in order to transfer analysis techniques of data obtained from field surveys and chemical analysis to the counterparts, the Central Department of Geography (CDG) at Tribhuvan University for three months (from September to November, 2016), one lecturer was invited to University of Yamanashi, and short-term training was provided. In addition, long-term training was conducted by inviting one CDG master's student from Tribhuvan University as a research student at University of Yamanashi (starting a doctoral course from April 2017) for four years from April 2016. From February 2017, joint instruction has been provided to one undergraduate student at Tribhuvan University. One short-term trainee was invited to University of Yamanashi in October-December 2017 and July-September 2018, and conducted data analysis of water quality and isotope analysis, writing a dissertation, and a series of activities required for technology transfer.

Working group 3 (Microbial and public health assessment)

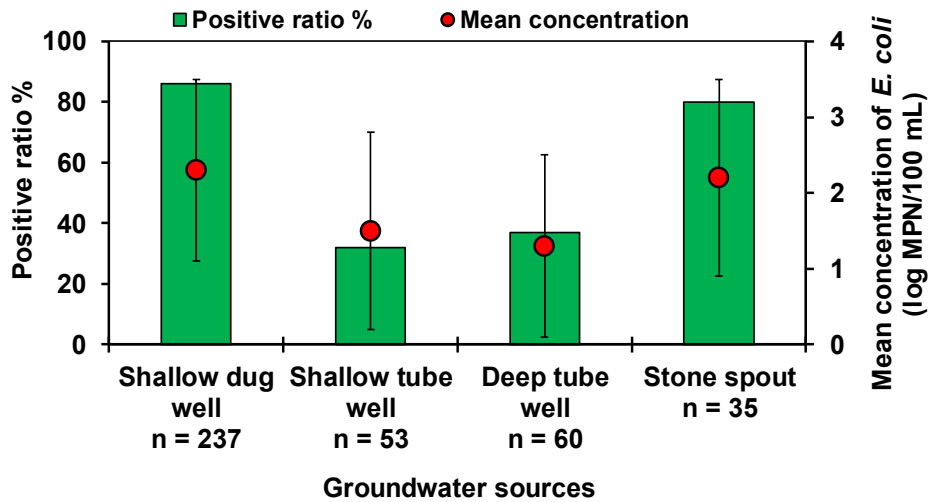
A.) Major results



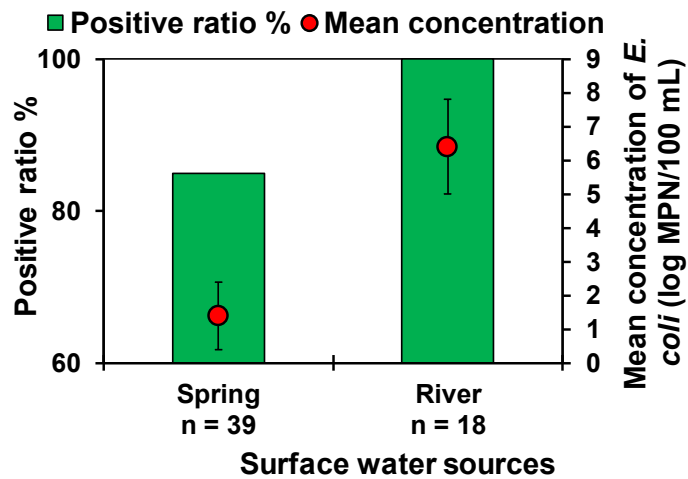
a. Distribution of *Escherichia coli* concentrations throughout the Kathmandu Valley (2016 Dry season)



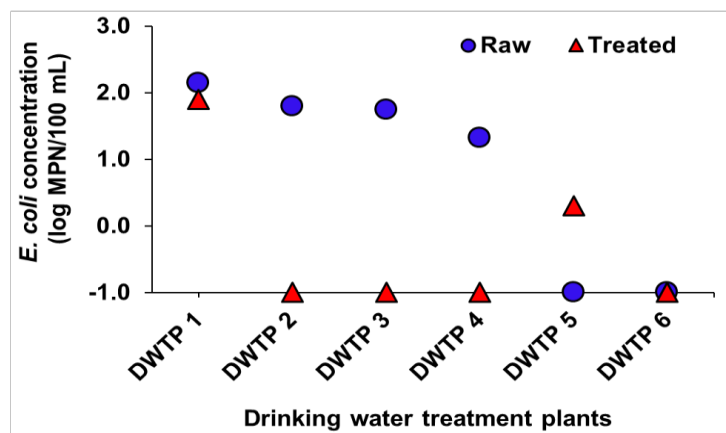
b. Distribution of *Escherichia coli* concentrations throughout the Kathmandu Valley (2016 Wet season)



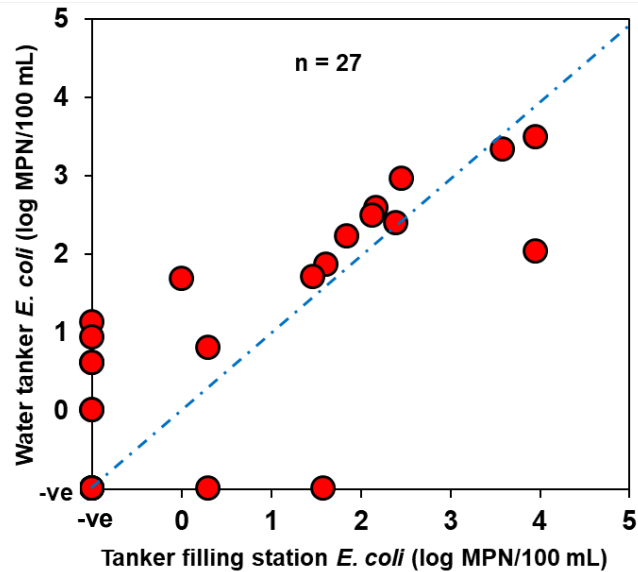
c. Positive ratio and detected concentrations of *E. coli* by type of groundwater



d. Positive rate and detected concentration of *Escherichia coli* in spring and river water



e. *E. coli* concentration in raw water and treated water of drinking water treatment plants



- f. Comparison of *E. coli* concentrations in water pumped into and drained out of a water tanker

B.) Status of technology transfer to the counter parts

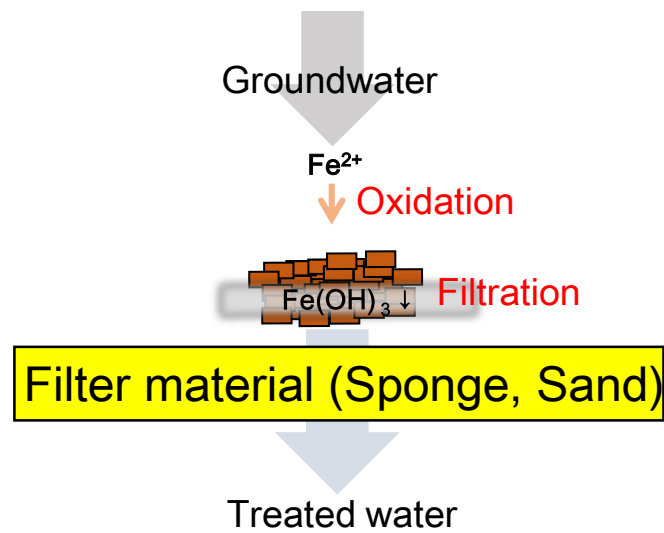
So far, field surveys were conducted 12 times (September 2014, July, August, and December 2015, February to March, June, and August 2016, December 2016 to January 2017, August to September 2017, April to May, June to July, and August 2018). The members of the microbiology laboratory, Institute of Medicine, Tribhuwan University, were provided technical guidance and training on laboratory methods. A total of four researchers got the opportunity to take training at the University of Yamanashi, Japan. In particular, during the large-scale water sampling surveys, conducted in February to March 2016 (dry season) and August 2016 (wet season), in cooperation with WG2 members, the counterpart members were the main actors and were able to perform laboratory works for microbial analysis of more than 300 water samples.

As a JICA short-term trainee, one senior researcher and two research staffs from Institute of Medicine, Tribhuwan University were accepted twice from October to November 2014 and another research staff twice from October to December 2017 and July to August 2018. They were provided training on techniques for the detection and measurement methods of the microorganisms. A total of three researchers, one each in October 2015, 2016, and 2018 were accepted.

Working group 4 (Water treatment system development)

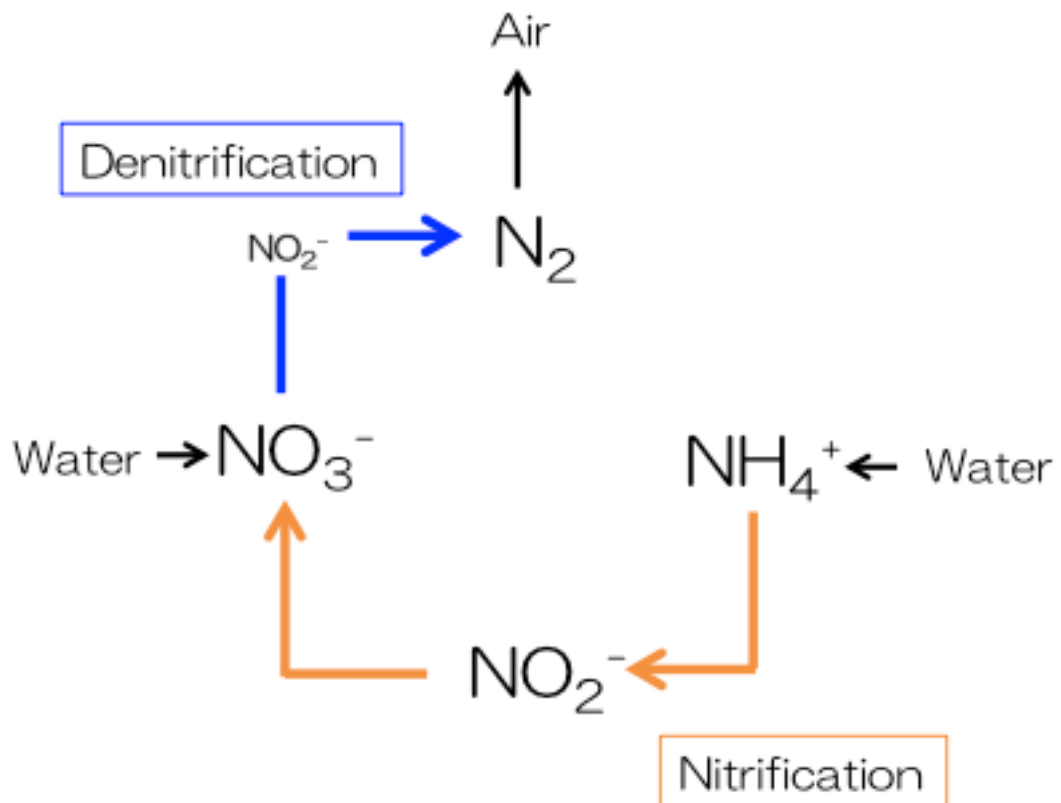
A.) Detailed mechanisms of the LCD system

Iron removal system



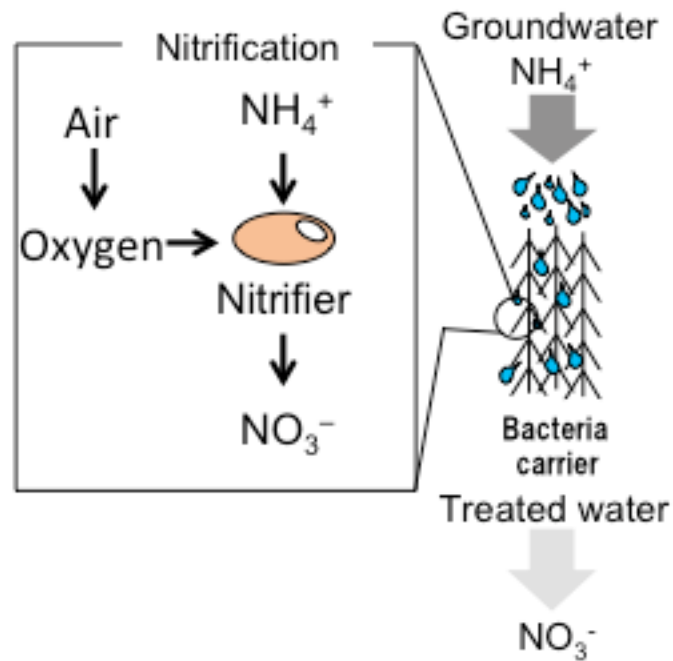
Schematic images of iron removal mechanism

Nitrogen removal system



Schematic diagram of nitrogen removal flow

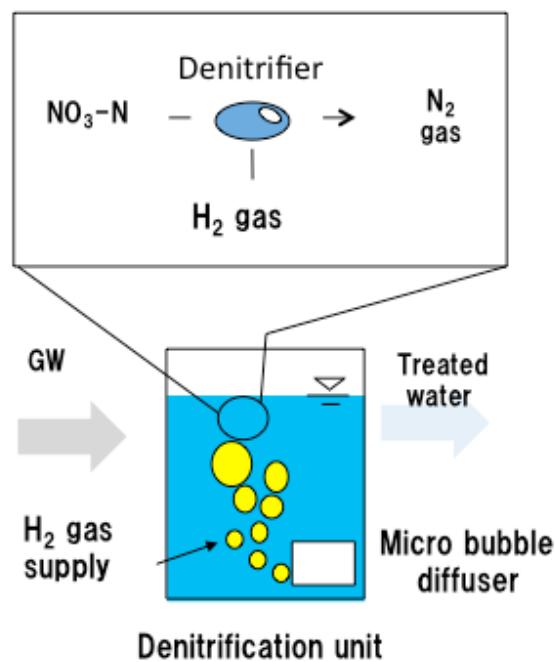
(1) Nitrification system for NH₄⁺ removal



Schematic image of basic concept of nitrification unit of LCD system

(2) Denitrification system for NO₃⁻ removal

Hydrogen depending denitrification by microbiological process



Schematic images of denitrification unit of LCD system

B.) Details of the installed LCD

Site 1: Jwagal (UN Park)

- LCD treatment system components: Sponge inclined water treatment device, dropping nitrification device, hydrogen oxidation denitrification device, activated carbon filtration device, anammox treatment device, artificial wetland system.
- Purpose: Performance evaluation of each device related to iron and nitrogen removal from groundwater, supply of domestic and drinking water.
- Capacity: 1000L/ day

Site 2: Chyasal

- LCD treatment system components: Dropping nitrifier, sand filter, activated carbon filter
- Purpose: Removal of ammonia nitrogen from raw water (shallow groundwater), supply of drinking water.
- Water supply target: Nearly 130 households
- Capacity: 3000 L/ day

Site 3: Chapacho (Thimi)

- LCD treatment system components: Hydrogen oxidation denitrification device, activated carbon filter
- Purpose: Removal of nitrate nitrogen from raw water (shallow groundwater), drinking water supply
- Water supply target: Nearly 100 households
- Capacity: 500 L/ day

Site 4: Institute of Engineering, Girls' Hostel

- LCD constituting device: Dropping nitrification device, sand filter device, activated carbon filter device
- Purpose: Removal of ammonia nitrogen and turbidity from raw water (deep groundwater), supply of shower water.
- Water supply target: About 60 students
- Capacity: 1500L/ day

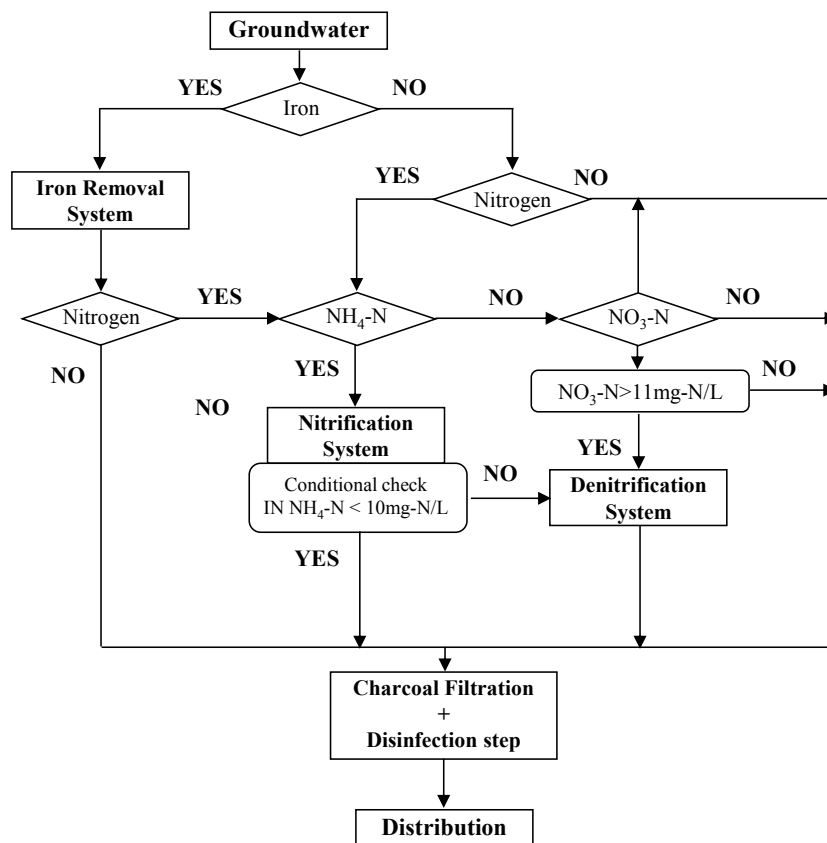
Site 5: KUKL water treatment plant in Lokanthali

- LCD component device: Dropping nitrification device, hydrogen denitrification device, activated carbon filter device
- Purpose: Nitrogen removal from raw water, drinking water supply
- Capacity: 1000L/ day

Site 6: Residence

- LCD component device: Sponge inclined water treatment device, sand filtration device
- Purpose: Iron removal from raw water, supply of washing water and shower water

C.) Selection of LCD system for drinking water distribution



D.) Status of technology transfer to counter part

In 2014, the laboratory, which is the research base for Nepal, was established at IOE, Tribhuvan University. In addition, in 2014, four JICA short-term trainees (engineer with IOE laboratory, Deputy Director of KUKL Water Quality Division, member of CREEW,

Master's student at Tribhuwan University) were invited to University of Yamanashi and instructed on water quality analysis technology for nitrogen treatment performance evaluation.

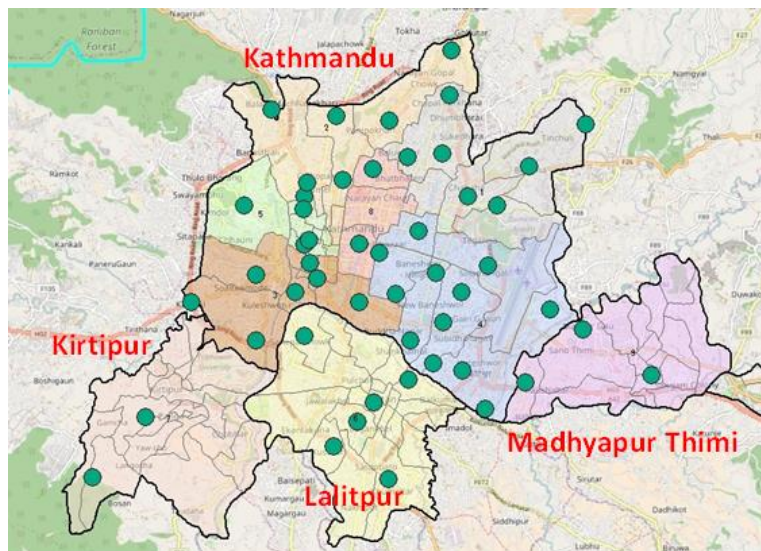
In 2015, 18 students from August 20-22, 2015, 11 students from IOE, Tribhuvan University and NGO staff from December 20-27 2015 were accepted by IOE laboratory, and trainings on water quality analysis were conducted. Technology transfer was carried out. In addition, water quality analysis texts were prepared for this training and distributed to the participants.

In 2016, four JICA short-term trainees (2016.9.5-11.30 IOE Master's 3 students and 3 staff members of KUKL's water quality field) were accepted to provide training on water quality analysis technology and nitrogen and iron water treatment technology. In 2017, three short-term research students from JICA (2017.10. 13-31, 2017.10.1-2017.12.22) were accepted and instructed on water quality analysis technology, LCD core technology, and LCD design.

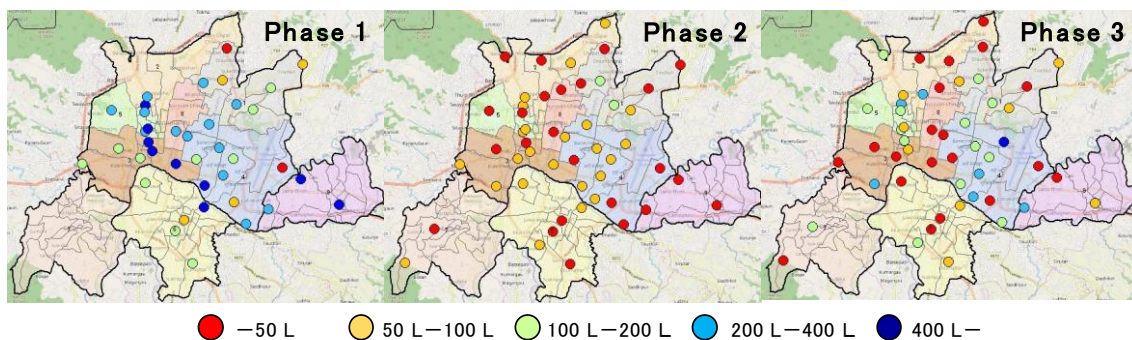
In 2018, three short-term research students from JICA were accepted to train on water quality analysis technology and on water treatment technology of nitrogen and iron.

Working group 5 (Socio-economic assessment)

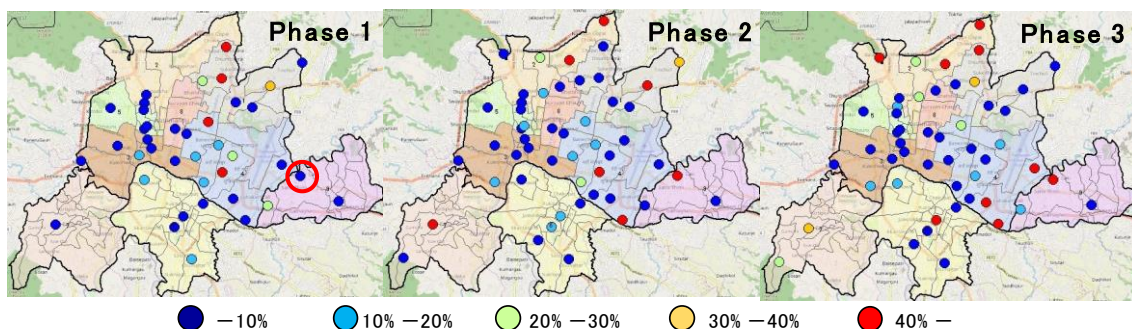
A.) Major results



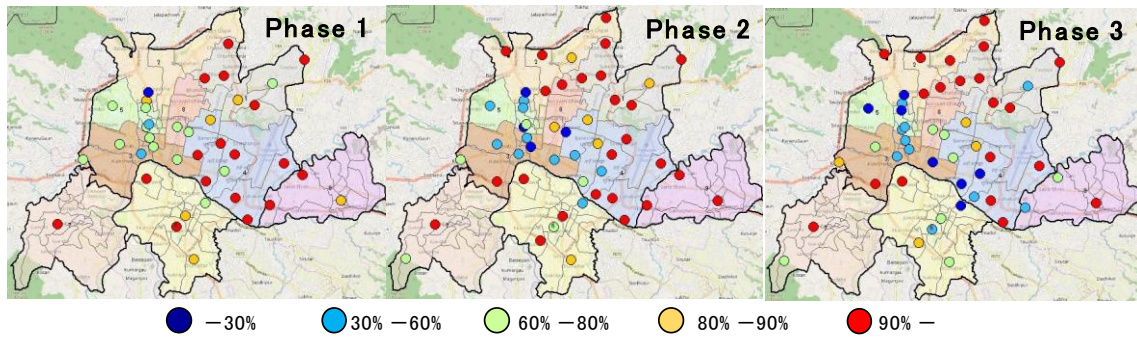
a. Locations of the clusters of household (N=1500) questionnaire survey



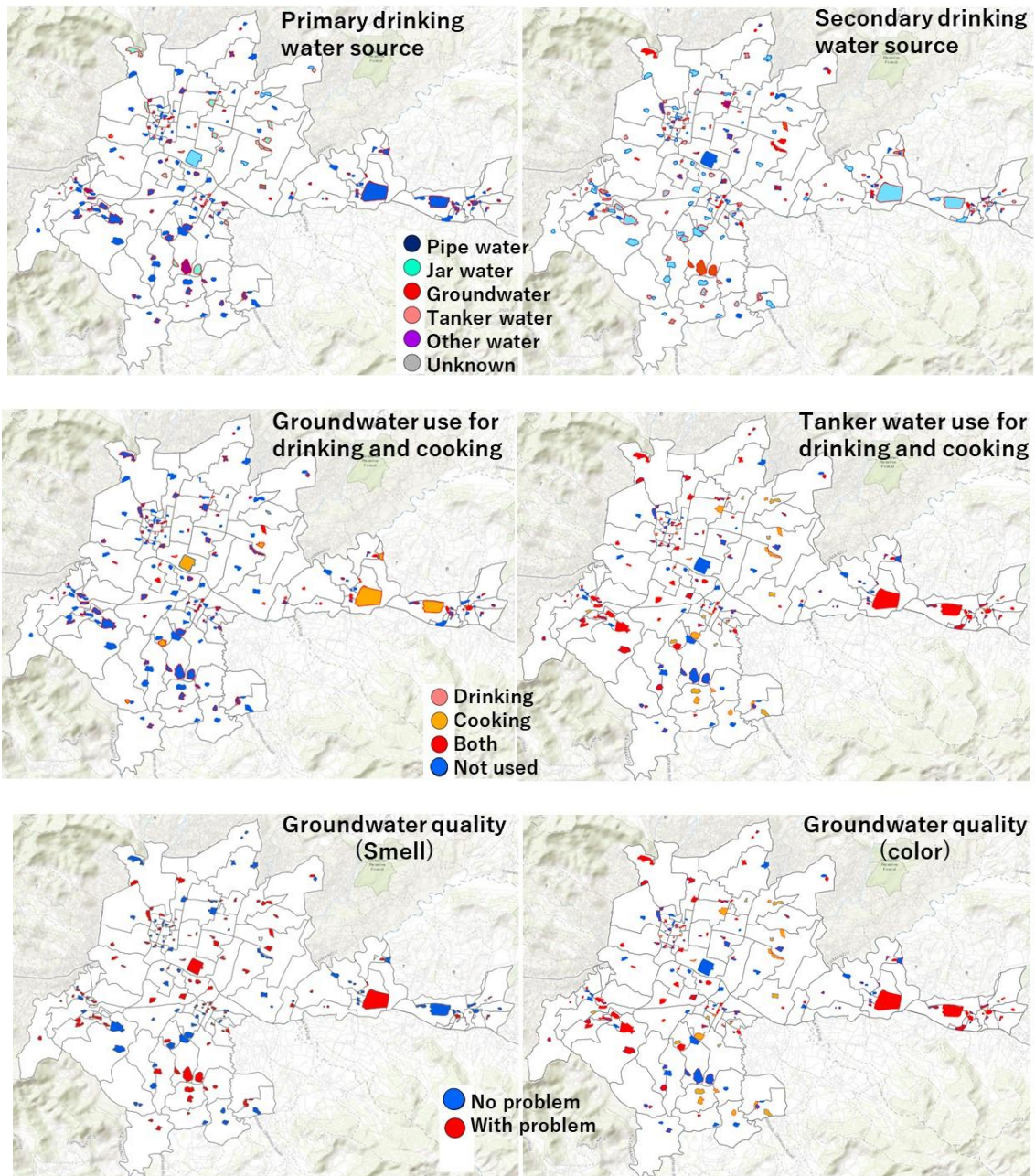
b. Average piped-water consumption in the Kathmandu Valley (unit: Litre per day in each cluster) (Phase I: 2015 dry season; Phase II: 2016 dry season; Phase III: 2016 wet season)



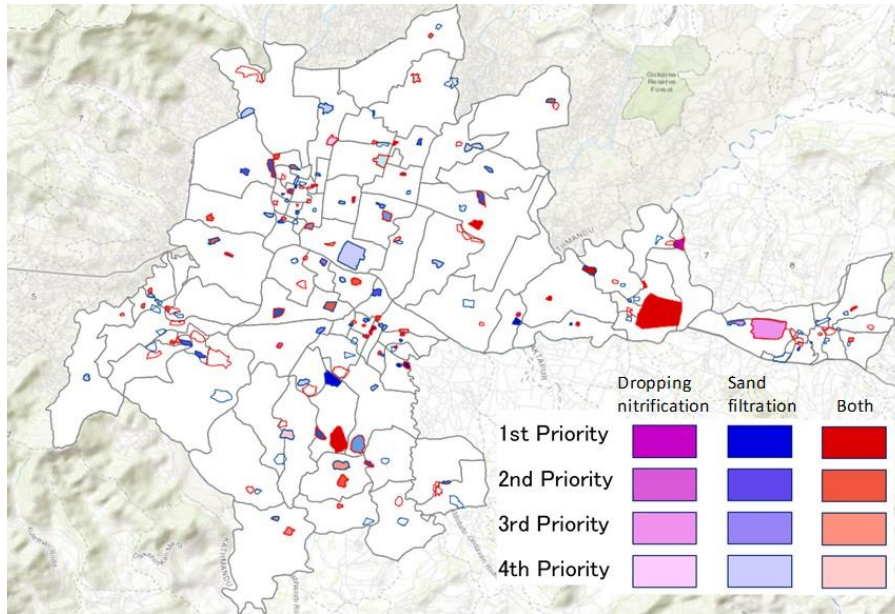
c. Ratio of households using groundwater for drinking and cooking



d. Ratio of households treating drinking water before use

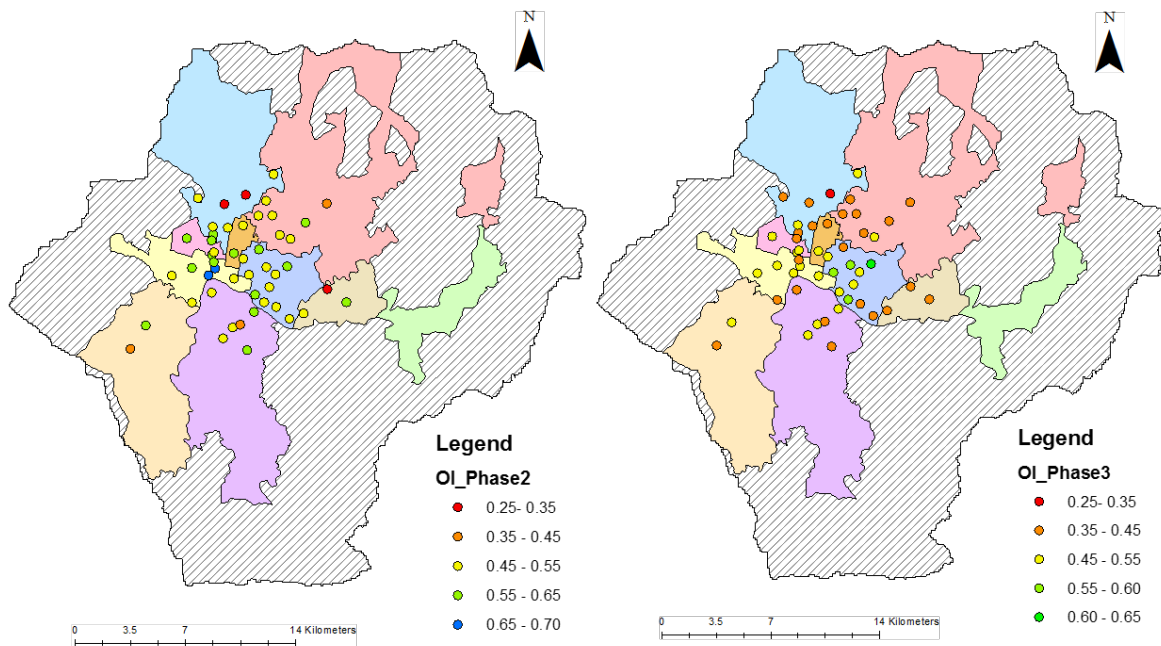


e. Water use in communities in all wards in central Kathmandu basin

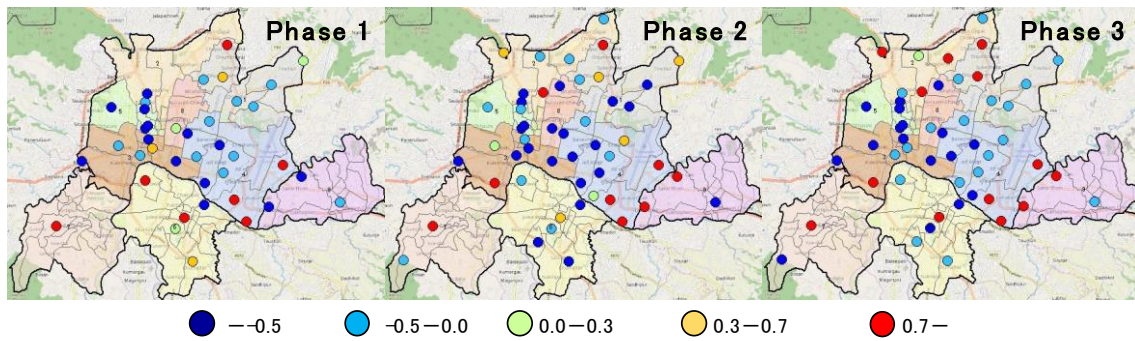


f. Candidate communities for LCD system installation in priority based on groundwater quality and ingested water

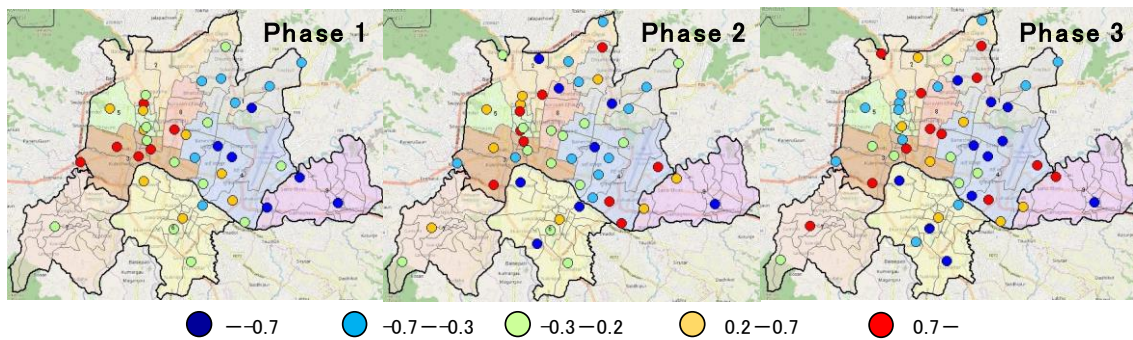
(1st: Drinking groundwater with poor quality, 2nd: Drinking tanker water with poor quality, 3rd: Drinking tanker water without quality problem, 4th: Drinking Jar water)



g. Objective index (OI) of water security in the municipal areas during Phase 2 and Phase 3



h. Water stress felt by the residents (Inconvenience in daily life)



i. Water stress felt by the residents (Anxiety to water safety)

B.) Status of technology transfer to the counterpart

From 2014 to 2016, we improved the question items and the survey method regarding the questionnaire survey, and conduct detailed meetings with the counterpart regarding the method of securing the continuity of the survey, and also for the surveyor when Japanese members visited Nepal. The trainings were conducted to transfer and acquire methods. In addition, we made manual about contents of household investigation and area investigation every word and investigation method and provided to the counterpart (September, 2018).

From 2016 (2016.4-), one member from the Tribhuvan University and one member from the Ministry of Urban Development entered the doctoral course of our university as a government funded foreign student. In 2017 and 2018, we received one short-term trainee and provided technical guidance on the construction and application of a regional scale economic model.

III.) List of the Peer-reviewed Papers and Joint Academic Presentations

WG 1

Peer-reviewed Papers: 7 (Individual: 0)

1. Thapa, B.R., Ishidaira, H., Pandey, V.P. and Shakya, N.M.: Impact assessment of Gorkha Earthquake 2015 on potable water supply in Kathmandu Valley: Preliminary analysis, Journal of Japan Society of Civil Engineering, 2015, Ser. B1 (Hydraulic Engineering), 72, 4, I_61-I_66
2. Udmale P., Ishidaira H., Thapa B. R., and Shakya. N.M.: The Status of Domestic Water Demand: Supply Deficit in the Kathmandu Valley, Nepal, Water, 2016, 8 (5), 196
3. Thapa, B.R., Ishidaira, H., Bui, T.H. and Shakya, N.M.: Evaluation of water resources in mountainous region of Kathmandu Valley using high resolution satellite precipitation product, Journal of Japan Society of Civil Engineering, Ser. G (Environment), 2016, 72 (5), I_27-I33.
4. Thapa, B.R., Ishidaira, H., Pandey, V.P. and Shakya, N.M.: A multi-model approach for analyzing water balance dynamics in Kathmandu Valley, Nepal, Journal of Hydrology: Regional Studies, 2016, 9, 149–162
5. Bhesh Raj Thapa, Hiroshi Ishidaira, Vishnu Prasad Pandey, Tilak Mohan Bhandari and Narendra Man Shakya : Evaluation of Water Security in Kathmandu Valley before and after Water Transfer from another Basin, Water, 2018, 10 (2), 224
6. Aryal A., Magome J., Pudashine J. R., Ishidaira H.: Identifying the potential location of hydropower sites and estimating the total energy in Bagmati river basin, Journal of Japan Society of Civil Engineering, Ser. G (Environment), Vol.74, No.5, I_315-I_321; 10.2208/jscejer.74.I_315
7. Bhesh Raj Thapa, Hiroshi Ishidaira, Maksym Gusyev, Vishnu Prasad Pandey, Parmeshwar Udmale, Masaki Hayashi, Narendra Man Shakya: Implications of the Melamchi Water Supply Project for the Kathmandu Valley groundwater system. Water Policy, 2018 accepted

Joint Academic Presentations: 8

1. Thapa, B. R., Ishidaira, H., Gusev, M., Pandey, V. P., Udmale, P., Hayashi, M., and Shakya, N. M. , Implications of Melamchi Water Supply Project in the groundwater resources of the Kathmandu Valley, Ground water Symposium in Nepal, March 20, Kathmandu, Nepal.
2. Thapa Bhesh Raj, Ishidaira Hiroshi, Pandey Vishnu Prasad, Udmale P, Hayashi M, Gusev M, Shakya N.M: 2016.Groundwater management issues in Kathmandu Valley after Melamchi Water Supply Project (MWSP), COPING AND COMPLEXITY:Maximising Public Value from Kathmandu's Melamchi Water Supply Project, 2016.9.15-16
3. Bhesh Raj Thapa, Hiroshi Ishidaira, Narendra Man Shakya: The status of domestic water supply on pre and post Melamchi water supply project in Kathmandu Valley, Nepal, 5th International Youngresearchers' Workshop on River Basin Environment and Management, Ho Chi Minh, Vietnam, 2016.11.12-13
4. Thapa, B. R., Ishidaira, H., Pandey, V. P., Bhandari T.M., and Shakya, N. M. , Water Security Perspective On Pre And Post Melamchi Water Supply Project In Kathmandu Valley, Nepal, Water Security And Climate Change: Challenges And Opportunities In Asia, 29 November-01 December, 2016, Asian Institute of Technology, Bangkok, Thailand
5. Basnet S., Shakya N.M., Ishidaira H.: Simulation of Kathmandu valley river basin hydrologic process using coupled ground and surface water model, WATER SECURITY AND CLIMATE CHANGE:CHALLENGES AND OPPORTUNITIES IN ASIA, 29 November-01 December, 2016, Asian Institute of Technology, Bangkok, Thailand
6. Kozono T, Bhesh R Thapa, Ishidaira H: Estimating agricultural water demand in the Kathmandu Valley using CROPWAT model and satellite observations, 3rd International Young Researchers Workshop on River Basin Environment and Management, Phitsanulok, Thailand, 2015.12.21-22
7. 小園智皓、石平博、ベシユ ラジ タパ : リモート・センシング技術を利用した農作物分布特定と農業用水需要量の推計、日本地球惑星科学連合 2016 年大会、千葉、2016.5.20-25
8. 原佑太郎、馬籠純、石平博、Bhesh Raj THAPA、Hieu Thi BUI: カトマンズ盆地における屋根雨水利用ポテンシャルの評価。水文・水資源学会 2017 年度研究発表会 北見 2017.9.20

WG 2

Peer-reviewed Papers: 3 (Individual: 2)

1. Sadhana Shrestha, Takashi Nakamura, Rabin Malla, Kei Nishida: Seasonal variation in the microbial quality of shallow groundwater in the Kathmandu Valley, Nepal. *Water Science and Technology*, 2014, 14 (3), 390-397.
2. Rabin Malla, Sarita Shrestha, Saroj K. Chapagain¹, Maneesha Shakya, Takashi Nakamura : Physico-Chemical and Oxygen-Hydrogen Isotopic Assessment of Bagmati and Bishnumati Rivers and the Shallow Groundwater along the River Corridors in Kathmandu Valley, Nepal, *Journal of Water Resource and Protection*, 2015, 7, 1435-1448.
3. Ramita Bajracharya, Takashi Nakamura, Bijay Man Shakya, Kei Nishida, Suresh Das Shrestha, Naresh Kazi Tamrakar: Identification of river water and groundwater interaction at central part of the Kathmandu valley, Nepal using stable isotope tracers, *International Journal of Advanced Scientific and Technical Research*. 2018, 8(3), 29-4.
4. 中村 高志、西田 継、風間 ふたば、尾坂 兼一、Saroj K. Chapagain. ネパール・カトマンズ盆地における浅層地下水の窒素汚染. *日本水文学会誌*. 2014. 44(4):197-206
5. Ramita Bajracharya, Naresh Kazi Tamrakar, Manish Shrestha, and Bimal Bohara: Status of shallow wells along major rivers of the Kathmandu Valley, Central Nepal, *Journal of Nepal Geological Society*, 56, 31-42.

Joint Academic Presentations: 13

1. A. Khanal, S.D.Shrestha, T. Nakamura, M. Rijal, K. Nishida and S. Shrestha, Shallow aquifer characterization of southern part of Kathmandu Valley, Seventh Nepal Geological Congress, Kathmandu, 2015.4.7
2. K. Anoj, S.D.Shrestha, T. Nakamura, M. Rijal, K. Nishida and S. Shrestha, Shallow aquifer characterization of southern part of Kathmandu. 6th National Groundwater Symposium, 2015.
3. Takashi Nakamura, Kei Nishida, Yuki Yamamoto, Kodai Hiraga, Anoj Khanal, Shresh das Shrestha, Futaba Kazama, Ammonium sources of groundwater in Kathmandu Valley, Nepal, Japan Geoscience Union Meeting 2015 Chiba 2015.5.27
4. Takashi Nakamura, Kei Nishida, Suresh Das Shrestha, Yuki Yamamoto, Kazuki Akahane, Yasuhiro Takimoto : Ammonium and nitrate contamination source and dynamics in groundwater of Kathmandu Valley, Nepal. Japan Geoscience Union Meeting 2018 Chiba 2016.5.20-25.
5. Takashi Nakamura, Bijay Man Shakya, Eiji Haramoto, Kei Nishida, Jeevan B. Sherchand, Suresh Das Shrestha : Water quality and hydrological characteristic of mountain spring water on Kathmandu Valley, Nepal ~Alternative water resources on the event of a disaster~: Japanese Association of Hydrological Science 2016 Conference, Tokyo, 2016.Oct.15.
6. Bijay Man Shakya, Takashi Nakamura, Suresh Das Shrestha, Ramita Bajracharya, Kei Nishida: Source identification of serious ammonium contamination in groundwater of Kathmandu Valley: using stable isotope tracer technique. The 5th International Young Researchers Workshop on River Basin Environment and Management, Hotel Swiss Garden Resort, Kuantan, Malaysia, 2017.10.28-29
7. Masanari Morita, Shakya Bijay, Shrestha Suresh, Takashi Nakamura, Kei Nishida: Analysis of Nitrogen Pollution of Ground water in Kathmandu Valley. Japan Geoscience Union Meeting 2018 Chiba 2018.5.23
8. Bijay Man Shakya, Takashi Nakamura, Suresh Das Shrestha, Kei Nishida: Identification of the deep groundwater recharge process in Kathmandu Valley, Nepal. Japan Geoscience Union Meeting 2018 Chiba 2018.5.23
9. Bijay Man Shakya, Takashi Nakamura, Suresh Das Shrestha, Ramita Bajracharya, Kei Nishida: Identification of nitrogen contamination sources in shallow and deep groundwater of Kathmandu Valley, Nepal using stable isotope technique. International Conference on Water, Environment and Climate Change: Knowledge sharing and partnership 2018 Kathmandu, Nepal 2018.4.10-12
10. Ramita Bajracharya, Takashi Nakamura, Naresh Kazi Tamrakar, Bijay Man Shakya, kei Nishida, and Subesh Ghimire: Spatial variation of stable isotope and chemical concentration in groundwater and river water of the Kathmandu Valley. 9th Geological Congress, Kathmandu, 2018.
11. Ramita Bajracharya, Takashi Nakamura, Bijay Man Shakya, Kei Nishida, Suresh Das Shrestha and Naresh Kazi Tamrakar : Identification of river water and groundwater interaction at central part of the Kathmandu valley, Nepal. International Conference on Water, Environment and Climate

- Change: Knowledge sharing and partnership 2018 Kathmandu, Nepal 2018.
12. Bijay Man Shakya, Takashi Nakamura, Suresh Das Shrestha, Kei Nishida: Identifying the spatial distribution of the deep groundwater recharge processes using hydrogeochemical and stable isotope of water. The 10th National Groundwater Symposium Challenges and Opportunities for Sustainable Groundwater Resources Management in Nepal 2019 Kathmandu 2019.3.18
 13. Takashi Nakamura, Kei Nishida, Mohamad Naim, Shakya Man Bijay, Kazuyoshi Asai, Suresh Das Shrestha: Identification of deep groundwater recharge system in an intermontane basin. Japan Geoscience Union Meeting 2019 Chiba 2019.5.27 (accepted)

WG 3

Peer-reviewed Papers: 14 (Individual: 0)

1. Daisuke Inoue, Takuji Hinoura, Noriko Suzuki, Junqin Pang, Rabin Malla, Sadhana Shrestha, Saroj Kumar Chapagain, Hiroaki Matsuzawa, Takashi Nakamura, Yasuhiro Tanaka, Michihiko Ike, Kei Nishida, Kazunari Sei :High-throughput DNA microarray detection of pathogenic bacteria in shallow well groundwater in the Kathmandu Valley, Nepal. *Current Microbiology*, 2015, 70, 43-50
2. Sadhana Shrestha, Eiji Haramoto, Rabin Malla, Kei Nishida: Risk of diarrhoea from shallow groundwater contaminated with enteropathogens in the Kathmandu Valley, Nepal. *Journal of Water and Health*, 2014, 13 (1), 259-269
3. Bikash Malla, Rajani Ghaju Shrestha, Dinesh Bhandari, Sarmila Tandukar, Sadhana Shrestha, Hayato Yoshinaga, Daisuke Inoue, Kazunari Sei, Kei Nishida, Yasuhiro Tanaka, Jeevan B. Sherchand, Eiji Haramoto: Detection of *Cryptosporidium*, *Giardia*, fecal indicator bacteria, and total bacteria in commercial jar water in the Kathmandu Valley, Nepal, *Journal of Institute of Medicine*, 2014, 37(2):10-15.
4. Dinesh Bhandari, Sarmila Tandukar, Eiji Haramoto, and Jeevan B. Sherchand. 2016. Determination of fecal indicator bacteria in shallow and deep groundwater sources in the Kathmandu valley, Nepal. *Naresuan University Engineering Journal*. 11(1), 43-46.
5. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla, Dinesh Bhandari, Sarmila Tandukar, Daisuke Inoue, Kazunari Sei, Jeevan B. Sherchand, Eiji Haramoto, Next-generation sequencing identification of pathogenic bacterial genes and their relationship with fecal indicator bacteria in different water sources in the Kathmandu Valley, Nepal. *Sci Total Environ*. 2017 Dec 1; 601-602:278-284.
6. Sadhana Shrestha, Shankar Shrestha, Junko Shindo, Jeevan B. Sherchand, Eiji Haramoto, Virological quality of irrigation water sources and pepper mild mottle virus and tobacco mosaic virus as index of pathogenic virus contamination level *Food Environ Virol*. 2018 Mar; 10(1):107-120.
7. Sarmila Tandukar, Jeevan B. Sherchand, Dinesh Bhandari, Samendra P. Sherchan, Bikash Malla, Rajani Ghaju Shrestha, and Eiji Haramoto: Presence of human enteric viruses, protozoa, and indicators of pathogens in the Bagmati River, Nepal. *Pathogens*. 7(2):38. 2018.4.
8. Daisuke Inoue, Hayato Yoshinaga, Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Dinesh Bhandari, Yasuhiro Tanaka, Jeevan B. Sherchand, Eiji Haramoto, and Kazunari Sei: Comprehensive detection of pathogenic bacteria in jar water, community well groundwater, and environmental water in the Kathmandu Valley, Nepal. *Japanese Journal of Water Treatment Biology*. 54(2):65-72. 2018.6.
9. Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Yasuhiro Tanaka, Jeevan B. Sherchand, and Eiji Haramoto: Validation of host-specific Bacteroidales quantitative PCR assays and their application to microbial source tracking of drinking water sources in the Kathmandu Valley, Nepal. *Journal of Applied Microbiology*. 125(2):609-619. 2018.8.
10. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Jeevan B. Sherchand, and Eiji Haramoto: Development of a quantitative PCR assay for *Arcobacter* spp. and its application to environmental water samples. *Microbes and Environments*. 33(3):309-316. 2018.9.
11. Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Yasuhiro Tanaka, Jeevan B. Sherchand, Eiji Haramoto. Identification of human and animal fecal contamination in drinking water sources in the Kathmandu Valley, Nepal, using host-associated Bacteroidales quantitative PCR assays. *Water*. 10(12):1796, 2018.12.
12. Sarmila Tandukar, Jeevan B. Sherchand, Surendra Karki, Bikash Malla, Rajani Ghaju Shrestha, Dinesh Bhandari, Ocean Thakali, Eiji Haramoto. Co-infection by waterborne enteric viruses in children with gastroenteritis in Nepal. *Healthcare*. 7(1):9, 2019.01.

13. Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Jeevan B. Sherchand, Eiji Haramoto. Performance evaluation of human-specific viral markers and application of pepper mild mottle virus and crAssphage to environmental water samples as fecal pollution markers in the Kathmandu Valley, Nepal. Food and Environmental Virology. In press.
14. Rajani Ghaju Shrestha, Kazuko Sawada, Daisuke Inoue, Hayato Yoshinaga, Bikash Malla, Sarmila Tandukar, Dinesh Bhandari, Yasuhiro Tanaka, Kazunari Sei, Jeevan B. Sherchand, Eiji Haramoto. Comparison of pathogenic bacteria in water and fecal-source samples in the Kathmandu Valley, Nepal, using high-throughput DNA microarray. Biomedical Journal of Scientific & Technical Research. in press

Joint Academic Presentations: 25

1. Bikash Malla, Rajani Ghaju (TU-IOM), Dinesh Bhandari (TU-IOM), Sarmila Tandukar (TU-IOM), Takashi Furuya, Sadhana Shrestha, Hayato Yoshinaga, Daisuke Inoue, Kazunari Sei (Kitasato Univ), Kei Nishida, Yasuhiro Tanaka (UY), Jeevan B. Sherchand (TU-IOM), Eiji Haramoto (UY), Prevalence of Cryptosporidium, Giardia, multidrug-resistant Acinetobacter, and indicator bacteria in jar water in the Kathmandu Valley, Nepal, 2nd International Young Researchers' Workshop on River Basin Environment and Management, Hanoi, 2015.01.05.
2. Hayato Yoshinaga, Daisuke Inoue (Kitasato Univ), Bikash Malla (UT), Rajani Ghaju, Dinesh Bhandari, Sarmila Tandukar (TU-IOM), Yasuhiro Tanaka (UY), Jeevan B. Sherchand (TU-IOM), Eiji Haramoto (UY), Kazunari Sei (TU-IOM): Comprehensive analysis of pathogenic bacteria occurred in jar water, community well groundwater and river water in the Kathmandu Valley, Nepal, Water and Environment Technology Conference 2015, Tokyo, 2015.08.06.
3. Bikash Malla (UY), Rajani Ghaju, Dinesh Bhandari, Sarmila Tandukar (TU-IOM), Sadhana Shrestha (UY), Hayato Yoshinaga, Daisuke Inoue, Kazunari Sei (Kitasato Univ), Kei Nishida, Yasuhiro Tanaka (UY), Jeevan B. Sherchand (TU-IOM), Eiji Haramoto (UY): Microbial analysis of jar water and community wells in the Kathmandu Valley, Nepal, Water and Environment Technology Conference 2015, Tokyo, 2015.08.06.
4. 吉永隼人, 井上大介 (北里大学), Bikash Malla, 田中靖浩 (山梨大学), Jeevan B. Sherchand (トリブワン大学), 原本英司 (山梨大学), 清和成 (北里大学) : ネパール・カトマンズ盆地の各種水試料中における病原性細菌汚染の実態調査, 日本水処理生物学会第 52 回大会, 北九州, 2015 年 11 月 12 日.
5. Dinesh Bhandari, Sarmila Tandukar (TU-IOM), Eiji Haramoto (UY), Jeevan B. Sherchand (TU-IOM): Identification of fecal indicator bacteria in shallow and deep groundwater sources in Kathmandu Valley, Nepal, 3rd International Young Researchers' Workshop on River Basin Environment and Management, Thailand, 2015.12.21.
6. Sarmila Tandukar, Jeevan B. Sherchand, Dinesh Bhandari (TU-IOM), Eiji Haramoto (UY): Detection of waterborne enteropathogens from river water sample to trace the source of contamination in Nepal, Water Microbiology Conference 2016, Chapel Hill, 2016.05.17.
7. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla (UY), Dinesh Bhandari, Sarmila Tandukar (TU-IOM), Daisuke Inoue, Kazunari Sei (Kitasato Univ), Jeevan B. Sherchand (TU-IOM), Eiji Haramoto (UY): Characterization of bacterial community by next generation sequencing in different sources of water in the Kathmandu Valley, Nepal, Water and Environment Technology Conference 2016, Tokyo, 2016.08.28.
8. Bikash Malla, Rajani Ghaju Shrestha (UY), Dinesh Bhandari, Sarmila Tandukar (Kitasato Univ), Hitoha Moriyama, Ryota Sugaya (UY), Daisuke Inoue, Kazunari Sei (Kitasato Univ), Yasuhiro Tanaka (UY), Jeevan B. Sherchand (TU-IOM), Eiji Haramoto (UY): Validation of host-associated Bacteroidales genetic markers and fecal pollution tracking of drinking water sources in the Kathmandu Valley, Nepal, Water and Environment Technology Conference 2016, Tokyo, 2016.08.28.
9. Sadhana Shrestha, Shankar Shrestha, Junko Shindo (UY), Jeevan B. Sherchand (TU-IOM), Eiji Haramoto (UY): Quantifying occurrence of and modelling risk from human pathogenic viruses in wastewater used for irrigation in vegetables farms, 5th Food and Environmental Virology Conference, Kusatsu, 2016.09.14.
10. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla, Dinesh Bhandari, Sarmila Tandukar, Daisuke Inoue, Kazunari Sei, Jeevan B. Sherchand, Eiji Haramoto: Development of a quantitative PCR assay for 16S rRNA gene of Arcobacter spp. and its application to different water sources in the Kathmandu Valley, Nepal. ASM Microbe 2017 ニューオーリンズ, 2017.6.2
11. Rajani Ghaju Shrestha, Kazuko Sawada, Daisuke Inoue, Bikash Malla, Sarmila Tandukar, Dinesh

- Bhandari, Yasuhiro Tanaka, Kazunari Sei, Jeevan B. Sherchand, Eiji Haramoto: Identification of pathogenic bacteria in fecal samples using DNA microarray analysis. Water and Environment Technology Conference 2017 札幌 2017.7.23
12. Sarmila Tandukar, Dinesh Bhandari, Bikash Malla, Rajani Ghaju Shrestha, Jeevan B. Sherchand, Eiji Haramoto: Investigation of waterborne protozoa and viruses in the Bagmati River, Nepal. Water and Environment Technology Conference 2017 札幌 2017.7.23
 13. Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Yasuhiro Tanaka, Jeevan B. Sherchand, Eiji Haramoto: Microbial source tracking of alternative drinking water sources in the Kathmandu Valley, Nepal, using Bacteroidales quantitative PCR assays. Water and Environment Technology Conference 2017 札幌 2017.7.23
 14. Daisuke Inoue, Hayato Yoshinaga, Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Dinesh Bhandari, Yasuhiro Tanaka, Jeevan B. Sherchand, Eiji Haramoto, Kazunari Sei: Comprehensive detection of pathogenic bacteria in jar water, community well groundwater, and environmental water in the Kathmandu Valley, Nepal. 7th International Water Association - Asia Pacific Regional Group (IWA-ASPIRE) Conference 2017 2017.9.12
 15. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Jeevan B. Sherchand, Eiji Haramoto: Quantitative PCR detection of 16S rRNA and *ciaB* genes of *Arcobacter* spp. in human and animal fecal source samples in the Kathmandu Valley, Nepal. The 5th International Young Researchers Workshop on River Basin Environment and Management, Hotel Swiss Garden Resort, Kuantan, Malaysia, 2017.10.28-29
 16. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Jeevan B. Sherchand, and Eiji Haramoto: Characterization of bacterial community and detection of 16S rRNA and *ciaB* genes of *Arcobacter* spp. in different water sources in the Kathmandu Valley, Nepal. International Conference on "Water, Environment and Climate Change: Knowledge Sharing and Partnership". Kathmandu, Nepal. 2018.4.11
 17. Sarmila Tandukar, Dinesh Bhandari, Bikash Malla, Rajani Ghaju Shrestha, Jeevan B. Sherchand, and Eiji Haramoto: Quantitation of enteric viruses in river water and at wastewater treatment plants in the Kathmandu Valley, Nepal. International Conference on "Water, Environment and Climate Change: Knowledge Sharing and Partnership". Kathmandu, Nepal. 2018.4.11.
 18. Ocean Thakali, Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Jeevan B. Sherchand, and Eiji Haramoto: Prevalence of fecal indicator bacteria and protozoa in various water sources in the Kathmandu Valley, Nepal. International Conference on "Water, Environment and Climate Change: Knowledge Sharing and Partnership". Kathmandu, Nepal. 2018.4.11.
 19. Sarmila Tandukar, Bikash Malla, Rajani Ghaju Shrestha, Dinesh Bhandari, Jeevan B. Sherchand, and Eiji Haramoto: Presence of enteric viruses and protozoa in different sources of water in the Kathmandu Valley, Nepal. ASM Microbe 2018. Atlanta, USA. 2018.6.8.
 20. Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Yasuhiro Tanaka, Jeevan B. Sherchand, and Eiji Haramoto: Detection of pathogenic viruses and fecal-source markers in tanker water and its source in the Kathmandu Valley, Nepal. ASM Microbe 2018. Atlanta, USA. 2018.6.8.
 21. Niva Sthapit, Bikash Malla, Rajani Ghaju Shrestha, Sarmila Tandukar, Ocean Thakali, Jeevan B. Sherchand, Eiji Haramoto, and Futaba Kazama: Prevalence of Shiga toxin-producing *Escherichia coli* (STEC) in river water and wastewater in the Kathmandu Valley, Nepal. Water and Environment Technology Conference 2018. Matsuyama. 2018.7.14.
 22. Rajani Ghaju Shrestha, Kazuko Sawada, Daisuke Inoue, Hayato Yoshinaga, Bikash Malla, Sarmila Tandukar, Dinesh Bhandari, Yasuhiro Tanaka, Kazunari Sei, Jeevan B. Sherchand, and Eiji Haramoto: Identification of pathogenic bacteria in water samples in the Kathmandu Valley, Nepal, using DNA microarray analysis. Water and Environment Technology Conference 2018. Matsuyama. 2018.7.14.
 23. Sarmila Tandukar, Jeevan B. Sherchand, Bikash Malla, Rajani Ghaju Shrestha, Dinesh Bhandari, Ocean Thakali, and Eiji Haramoto: Factors associated with co-infection of waterborne enteric viruses in diarrheal children. Water and Environment Technology Conference 2018. Matsuyama. 2018.7.15.
 24. Rajani Ghaju Shrestha, Yasuhiro Tanaka, Bikash Malla, Sarmila Tandukar, Dinesh Bhandari, Daisuke Inoue, Kazunari Sei, Jeevan B. Sherchand, and Eiji Haramoto: Characterization of bacterial community in fecal-source samples in the Kathmandu Valley, Nepal, using next-generation sequencing. IWA World Water Congress & Exhibition 2018. Tokyo. 2018.9.18
 25. Mai Nakano, Tatsuru Kamei, Bijay Man Shakya, Bikash Malla, Rajani Ghaju Shrestha, Yasuhiro Tanaka, Suresh Das Shrestha, Takashi Nakamura, Kei Nishida, Eiji Haramoto, Tadashi Toyama, and Futaba Kazama: Distribution of anaerobic ammonium oxidation (anammox) bacteria in

groundwater of Kathmandu Valley in Nepal. IWA World Water Congress & Exhibition 2018. Tokyo. 2018.9.19

WG 4:

Peer-reviewed Papers: 1 (Individual: 8)

1. Tatsuru Kamei, Dai Naitoh, Wilawan Khanitchaidecha, Futaba Kazama: Simultaneous removal of ammonium and nitrate by a combination of Anammox and hydrogenotrophic denitrification. *Journal of Water and Environment Technology*, 2015, 13, 2, 167-178
2. Toyama T., Nishimura N., Ogata Y., Sei K, Mori K., Ike M: Effects of planting *Phragmites australis* on nitrogen removal, microbial nitrogen cycling, and abundance of ammonia-oxidizing and denitrifying . *Environmental Technologymicroorganisms in sediments. Environmental Technology*, 2015, 37, 4, 478-485
3. Tatsuru Kamei, Sachi Shimizu, Yasuhiro Tanaka, Futaba Kazama: Anaerobic ammonium oxidation bacterial communities in long-term cultivated sludge: a comparison between mesophilic and psychrophilic conditions. *Japanese Journal of Water Treatment Biology*, 2016, 52, 1, 1-9
4. R. Eamrat, T. Mochizuki, T. Kamei, F. Kazama: Hydrogenotrophic Denitrification Activity under Intermittent Hydrogen Supply using Micro-Bubble System. *Naresuan University Engineering Journal*. 11(1):47-51.
5. R. Eamrat, Y. Tsutsumi, T. Kamei, W. Khanichaidecha, F. Kazama: Optimization of hydrogenotrophic denitrification behavior using continuous and intermittent hydrogen gas supply. *Journal of Water and Environment Technology*.
6. Tatsuru Kamei, Rawintra Eamrat, Kenta Shinoda, Yasuhiro Tanaka, Futaba Kazama 2018 Coupled anaerobic ammonium oxidation and hydrogenotrophic denitrification for simultaneous NH₄-N and NO₃-N removal. *Water Science and Technology*. <https://doi.org/10.2166/wst.2018.459>
7. R. Maharajan, I.M. Amatya, R.K.Sharm, T. Kamei, F.Kazama: Comparative Analysis of Hydrogenotrophic Denitrification in Up and Down Flow reactors. *WASH Journal*, 2019, 17, 25-29
8. D. R. Chitrakar, I.M. Amatya: Comparative Study of Crushed Glass and Sand as Filter Media in Rapid Water Filtration. . *WASH Journal*, 2019, Vo. 17, 39-45
9. M.S.Saud, I.M.Amatya: Hydrogenotrophic-Denitrification Efficiency Variation in Single Reactor and Series Reactors. *WASH Journal*, 2019, Vo. 17, 54-59

Joint Academic Presentations: 21

1. 亀井樹、望月智耶、Sarad Pathak (CREEW)、風間ふたば：ネパール・カトマンズ市内における簡易型窒素除去装置を用いた地下水浄化の検討、日本陸水学会 甲信越支部会 第40回支部会、長野県、2014.11.29-30
2. Tatsuru Kamei, Mai Nakano, Sarad Pathak (CREEW), Futaba Kazama: On site anammox bacteria cultivation for groundwater treatment. A case study in Kathmandu, Nepal. *Water and Environment Technology Conferene 2016, Tokyo, Japan*, 2016.08.28
3. Kamei Tatsuru, Sarada Pathak, Rawintra Wamrat, Yuya Tsutsumi, Kenta shinoda, Tadasi Tohyama, Futaba Kazama : Development of locally fitted, compact and decentralized (LCD) systems for portable water treatment in Kathmandu Valley, Nepal, 5th International Youngresearchers' Workshop on River Basin Environment and Management, Ho Chi Minh, Vietnam, 2016.11.12-13
4. 亀井樹、Sarad Pathak、篠田健太、中野麻衣、風間ふたば、ネパール国カトマンズにおける地下水からの嫌気性アンモニア酸化細菌の集積培養、第42回日本陸水学会甲信越支部会、小諸、2016.11.26
5. 亀井樹、Sarad Pathak、中野麻衣、篠田健太、田中靖浩、風間ふたば。ネパール国カトマンズ地域におけるAnammox菌群の集積培養。第51回日本水環境学会年会、熊本。2017.3.15-17.
6. Tatsuru KAMEI, Mai Nakano, Kenta Shinoda, Sarad Phatak, Yasuhiro Tanaka, Iswar Man Amatha, Tadashi Tohyama, Futaba Kazama: Start up of the Onsite and Experimental Scale NH₄-N Removal System for Nitrogen-contaminated Groundwater by Anaerobic Ammonium Oxidation Process (Anammox) in Kathmandu Valley, Nepal. *Water and Environment Technology Conference 2017 札幌* 2017.7.23
7. Mai Nakano, Tatsuru Kamei, Bikash Malla, Rajani Ghaju Shrestha, Yasuhiro Tanaka, Eiji Haramoto, Tadashi Toyama, Futaba Kazama: Distribution of anaerobic ammonium oxidation

- (anammox) bacteria in groundwater sources of Kathmandu Valley in Nepal. Water and Environment Technology Conference 2017 札幌 2017.7.23
8. Tatsuru Kamei, Sarad Pathak, Rawintra Eamrat, Yuya Tsutsumi, Kenta Shinoda, Tadashi Tohyama, Kazama Futaba: Development of Locally Fitted, Compact and Decentralized (LCD) Systems for Portable Water Treatment in Kathmandu Valley, Nepal. The 5th International Young Researchers Workshop on River Basin Environment and Management, Hotel Swiss Garden Resort, Kuantan, Malaysia, 2017.10.28-29
 9. Kenta Shinoda, Yuya Tsutsumi, Rawintra Wamrat, Tatsuru Kamei, Iswar Man Amatya and Futaba Kazama: Demonstration of Nitrate Removal by Hydrogenotrophic Denitrification. The 5th International Young Researchers Workshop on River Basin Environment and Management, Hotel Swiss Garden Resort, Kuantan, Malaysia, 2017.10.28-29
 10. 亀井 樹、篠田 健太、堤 裕也、中野 麻衣、Rawintra Emarat, 田中 靖浩, Iswal man Amatya, 遠山 忠、風間 ふたば: 小規模・自立分散型上水処理装置の開発-開発途上国ネパール・カトマンズ盆地の地下水浄化への適用- 日本水処理生物学会第 54 回大会 大阪 2017.11.9
 11. 亀井 樹、Sarad Pathak, 篠田健太、中野麻衣、堤裕也、Iswal man Amaty、田中 靖浩、遠山忠、風間 ふたば: 開発途上国での運用を想定した小規模・自立分散型上水処理装置の開発—ネパール・カトマンズ盆地における地下水浄化への適用—。第 54 回環境工学研究フォーラム 岐阜 2017.11.18
 12. 亀井 樹、Sarad Pathak, 篠田健太, Iswal man Amatya, 遠山忠, 風間ふたば: ネパール・カトマンズ盆地における地下水浄化装置開発とその性能評価. 日本陸水学会甲信越支部会第 43 回研究発表会 山梨県富士河口湖町 2017.11.26
 13. 中野麻衣, 亀井樹, Bikash Malla, Bijay Man Shakya, Rajani Ghaju Shrestha, 田中靖浩, Suresh Das Shrestha, 中村高志, 西田継, 原本英司, 遠山忠, 風間ふたば: ネパール国カトマンズ盆地における地下水源の嫌気性アンモニア酸化細菌 (Anammox 細菌) の分布と地質との関連性. 日本水処理生物学会第 54 回大会 大阪 2017.11.9
 14. 中野麻衣, 亀井樹, Bikash Malla, Rajani Ghaju Shrestha, Bijay Man Shakya, 田中靖浩, 中村高志, 西田継, Suresh Das Shrestha, 原本英司, 遠山忠, 風間ふたば: ネパール国カトマンズ地域における地下水源の嫌気性アンモニア酸化細菌(Anammox 菌)の分布. 第 54 回環境工学研究フォーラム 岐阜 2017.11.18
 15. 中野麻衣, 亀井樹, Bikash Malla, Rajani Ghaju Shrestha, Bijay Man Shakya, 田中靖浩, 中村高志, 西田継, Suresh Das Shrestha, 原本英司, 遠山忠, 風間ふたば: ネパール国カトマンズ盆地における地下水源中の嫌気性アンモニア酸化細菌(Anammox 細菌)の分布. 日本陸水学会甲信越支部会第 43 回研究発表会 山梨県富士河口湖町 2017.11.26
 16. 中野麻衣, 亀井樹, Bikash Malla, Bijay Man Shakya, Rajani Ghaju Shrestha, 田中靖浩, Suresh Das Shrestha, 中村高志, 西田継, 原本英司, 遠山忠, 風間ふたば: ネパール国カトマンズ盆地における嫌気性アンモニア酸化細菌(Anammox 細菌)の分布とその特徴. 第 52 回日本水環境学会年会 札幌 2018.3.17
 17. 亀井 樹、Sarad Pathak, 篠田健太、中野麻衣、堤裕也、Iswal man Amaty、田中 靖浩、遠山忠、風間 ふたば: 開発途上国ネパール・カトマンズ盆地での窒素汚染地下水の浄化—飲料水供給のための小規模・自立分散型上水処理装置の開発と適用—。第 52 回日本水環境学会年会 札幌 2018.3.17
 18. 篠田健太、堤裕也、Rawintra Eamrat、亀井樹、風間ふたば、Iswar Man Amatya: ネパール・カトマンズ盆地における水素酸化脱窒反応を用いた地下水中硝酸除去. 第 52 回日本水環境学会年会 札幌 2018.3.17
 19. Amit Kumar Maharjan, Do Hai Nam, Tadashi Toyama, Iswar Man Amatya, Futaba Kazama: Ammonium-Nitrogen Removal From Groundwater By Integrated Constructed Wetland. International Conference on "Water, Environment and Climate Change: Knowledge Sharing and Partnership". 2018.4.12
 20. Amit Kumar Maharjan, Do Hai Nam, Tadashi Toyama, Iswar Man Amatya, Futaba Kazama: Ammonium-nitrogen removal from groundwater by integrated constructed wetland reactor. Water and Environment Technology Conference 2018. Matsuyama. 2018.7.15.
 21. Tatsuru Kamei, Sarad Pathak, Kenta Shinoda, Iswar Man Amatya, Tadashi Toyama and Kazama Futaba: Feasibility analysis of locally fitted, compact and decentralized (LCD) system for water treatment from nitrogen contaminated groundwater. 6th International Young Researchers' Workshop on River Basin Environment and Management. Jimbaran Bay Beach Resort & Spa, Bali, Indonesia 2018.10.19-21

WG 5

Peer-reviewed Papers: 11 (Individual: 0)

1. Shankar Shrestha, Eiji Haramoto, Jeevan B. Sherchand, and Junko Shindo: Detection of coliform bacteria in irrigation water and on vegetable surfaces in the Kathmandu Valley of Nepal. *Journal of Institute of Medicine*, 2016, 38(1), 43-47.
2. Shankar Shrestha, Eiji Haramoto, Jeevan B. Sherchand, Sudarshan Rajbhandari, Meera Prajapati, and Junko Shindo. 2016. Seasonal variation of microbial quality of irrigation water in different sources in the Kathmandu Valley, Nepal. *Naresuan University Engineering Journal*. 11(1), 57-62.
3. Shankar Shrestha, Eiji Haramoto, Jeevan B. Sherchand, Sudeep Hada, Sudarshan Rajbhandari, and Junko Shindo. 2016. Prevalence of protozoa and indicator bacteria in wastewater irrigation sources in the Kathmandu Valley, Nepal: cases from Kirtipur, Bhaktapur and Madhyapur Thimi municipalities. *Journal of Water and Environment Technology*. 14(3), 149-157.
4. Sadhana Shrestha, Yoko Aihara, Arun Prasad Bhattarai, Niranjana Bista, Sudarshan Rajbhandari, Naoki Kondo, Futaba Kazama, Kei Nishida and Junko Shindo, Dynamics of domestic water consumption in the urban area of the Kathmandu Valley: Situation analysis pre and post 2015 Gorkha Earthquake, *Water* 2017, 9(3),222
5. Khadga Bahadur Shrestha, Bhesh Raj Thapa, Yoko Aihara, Sadhana Shrestha, Arun P. Bhattarai, Niranjana Bista, Futaba Kazama, Junko Shindo. Hidden Cost of Drinking Water Treatment and Its Relation with Socioeconomic Status in Nepalese Urban Context. *Water* 2018, 10, 607
6. Yoko Aihara, Sadhana Shrestha, Sudarshan Rajbhandari, Arun Prasad Bhattarai, Niranjana Bista, Futaba Kazama, Junko Shindo. Resilience in household water systems and quality of life after the earthquake: a mixed-methods study in urban Nepal, *Water Policy* wp2018117, 2018, doi.org/10.2166/wp.2018.117
7. Sadhana Shrestha, Yoko Aihara, Arun P. Bhattarai, Niranjana Bista, Naoki Kondo, Kazama Futaba, Kei Nishida, Junko Shindo. Development of an objective water security index and assessment of its association with quality of life in urban areas of developing countries. *SSM - Population Health* 6 (2018) 276–285.
8. Rajit Ojha, Bhesh Raj Thapa, Sadhana Shrestha, Junko Shindo, Hiroshi Ishidaira, Futaba Kazama: Water Price Optimization after the Melamchi Water Supply Project: Ensuring Affordability and Equitability for Consumer's Water Use and Sustainability for Utilities. *Water* 2018, 10, 249
9. Rajit Ojha, Bhesh Raj Thapa, Sadhana Shrestha, Junko Shindo, Hiroshi Ishidaira, Futaba Kazama; Water Taxation and Subsidy Analysis Based on Consumer Water Use Behavior and Water Sources Inside the Kathmandu Valley. *Water* 2018, 10(12), 1802
10. Shrestha, K.B.; Thapa, B.R.; Aihara, Y.; Shrestha, S.; Bhattarai, A.P.; Bista, N.; Kazama, F.; Shindo, J. Hidden 707 Cost of Drinking Water Treatment and Its Relation with Socioeconomic Status in Nepalese Urban Context. *Water* 2018, 10, 607
11. Shrestha, K.B, Kamei, T., Shrestha, S, Aihara, Y., Bhattarai, A.P., Bista, N., Thapa, B.R., Kazama, F. and Shindo, J.; Socioeconomic Impacts of LCD-Treated Drinking Water Distribution in an Urban Community of the Kathmandu Valley, Nepal, *Water* 2019, 11,1323

Joint Academic Presentations: 13

1. Shankar Shrestha, Eiji Haramoto (UY), Jeevan B. Sherchand (TU-IOM), Sudeep Hada, Sudarshan Rajbhandari (SEN), Junko Shindo (UY): Prevalence of protozoa and indicator bacteria in wastewater irrigation sources in the Kathmandu Valley, Nepal: cases from Kirtipur, Bhaktapur and Madhyapur Thimi municipalities, *Water and Environment Technology Conference 2015, Tokyo, 2015.08.05.*
2. Shankar Shrestha, Eiji Haramoto (UY), Jeevan B. Sherchand (TU-IOM), Sudarshan Rajbhandari (SEN), Meera Prajapati (CREEW), Junko Shindo (UY): Seasonal variation of microbial quality of irrigation water in different sources in the Kathmandu Valley, Nepal, 3rd International Young Researchers' Workshop on River Basin Environment and Management,
3. 稲垣達希、武藤慎一、新藤純子 (UY)、相原洋子 (KCCN)、Sudarshan Rajbhandari (SEN) : ネパール・カトマンズにおける生活の質 (QOL) に着目した水処理施設整備評価、土木学会関東支部第43回技術研究発表会、東京、2016.3.14-15.
4. Sadhana Shrestha (UY), Yoko Aihara (KCCN), Sudarshan Rajbhandari, Arun Prasad Bhattarai Niranjana Bista (SEN), Futaba Kazama, Junko Shindo (UY): Impact of 2015 Gorkha Earthquake on household water use and consequently on wellbeing in Kathmandu Valley, Nepal: A cohort study, *Asia Pacific Academic Consortium for Public Health Conference, Tokyo, 2016.9.16-19.*

5. Yoko Aihara (KCCN), Sadhana Shrestha (UY), Sudarshan Rajbhandari, Arun Prasad Bhattarai Niranjan Bista (SEN), Khadga Shrestha, Junko Shindo (UY): Resilience and recovery on water and health after 2015 Gorkha Earthquake, Nepal, Asia Pacific Academic Consortium for Public Health Conference, Tokyo, 2016.9.16-19.
6. 櫻田祥、武藤慎一、新藤純子、Sadhana Shrestha (UY)、Sudarshan Rajbhandari (SEN) : ネパール・カトマンズにおける地域別簡易水処理施設の整備評価、土木学会関東支部第 44 回技術研究発表会、埼玉、2017.3.7-8.
7. Sadhana Shrestha, Yoko Aihara, Arun Prasad Bhattarai, Niranjan Bista, Sudarshan Rajbhandari, Naoki Kondo, Kei Nishida, Futaba Kazama, Junko Shindo: Vulnerability of Poor Urban Women to Household Water Insecurity in Lalitpur Sub-Metropolitan City, Nepal. Gender Summit 10 東京, 2017.5.25
8. Khadga Bdr Shrestha, Arun Prasad Bhattarai, Nirajan Bista, Yoko Aihara, Sadhana Shrestha, Futaba Kazama, Junko Shindo: Social and economic determinants of drinking water treatment in the household of Kathmandu Valley. Water and Environment Technology Conference 2017 札幌 2017.7.23
9. Sadhana Shrestha, Yoko Aihara, Arun P. Bhattarai, Nirajan Bista, Sudarshan Rajbhandari, Naoki Kondo, Futaba Kazama, Kei Nishida, Junko Shindo: Association between quality of life and water insecurity in urban area of low-income country. 29th Annual Scientific Conference of the International Society of Environmental Epidemiology (ISEE17) シドニー 2017.9.26
10. Khadga Bahadur Shrestha, Arun Prasad Bhattarai, Niranjan Bista, Yoko Aihara, Sadhana Shrestha, Futaba Kazama, Junko Shindo, Hidden Cost for Drinking Water Treatment in Households of Kathmandu. International Conference on "Water, Environment and Climate Change: Knowledge Sharing and Partnership". Kathmandu, Nepal. 2018.4.11.
11. Takaaki Suzuki, Sadhana Shrestha, Arun Prasad Bhattarai, Nirajan Bista, Kei Nishida, Futaba Kazama, Hiroshi Ishidaira, Junko Shindo, Water insecurity perception affected by water availability and daily water use practice in Kathmandu Valley. Water and Environment Technology Conference 2018. Matsuyama. 2018.7.14.
12. Rajit Ojha, Bheshraj Thapa, Sadhana Shrestha, Junko Shindo, Futaba Kazama: Environmental externalities considerations in water prices of different options inside Kathmandu Valley. IWA World Water Congress & Exhibition 2018. Tokyo. 2018.9.19.
13. Khadga Bahadur Shrestha, Arun Prasad Bhattarai, Niranjan Bista, Junko Shindo: Water management situation of the community operated water supply systems in the Kathmandu Valley, Nepal. 6th International Young Researchers' Workshop on River Basin Environment and Management. Jimbaran Bay Beach Resort & Spa, Bali, Indonesia 2018.10.19-2

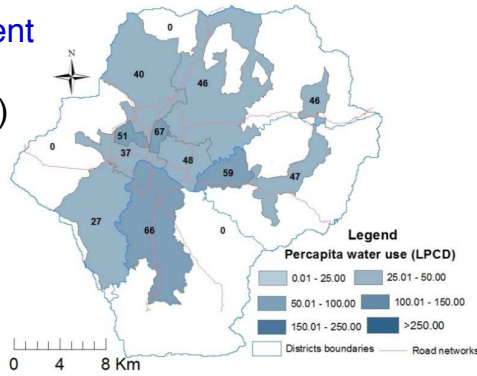
WATER SECURITY MAP



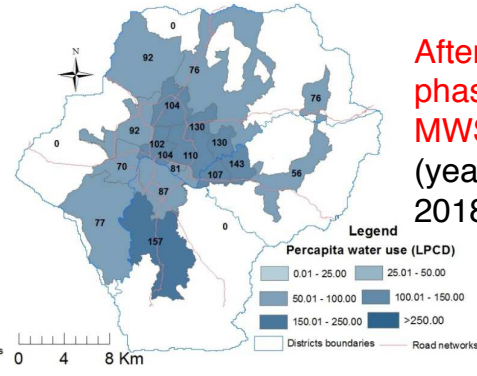
Water Security Map (WG1)

Spatial Distribution of Water supplied per capita (LPCD)

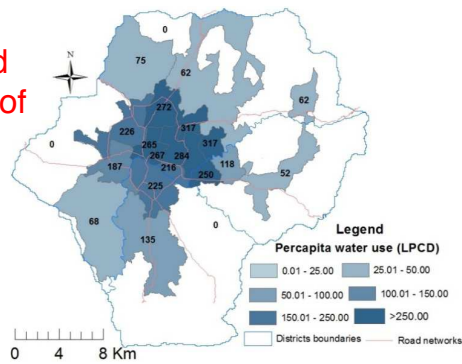
Current
(year 2016)



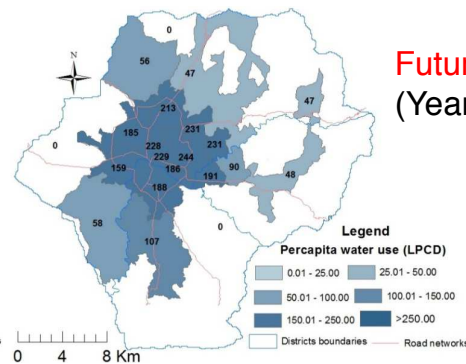
After first
phase of
MWSP
(year 2018)



After
Second
Phase of
MWSP
(Year 2023)

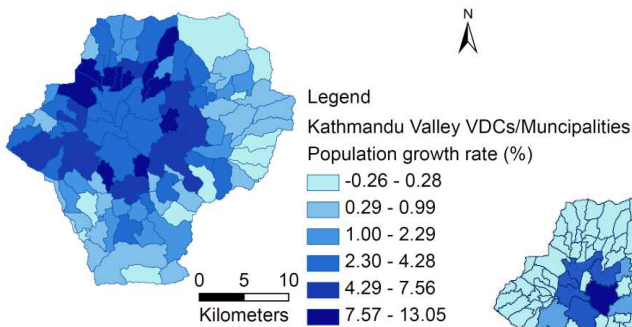


Future
(Year 2030)



Water Security Map (WG1)

Spatial Distribution of Water Demand



Growth ratio

$$P_t = P_0 * e^{rt}$$

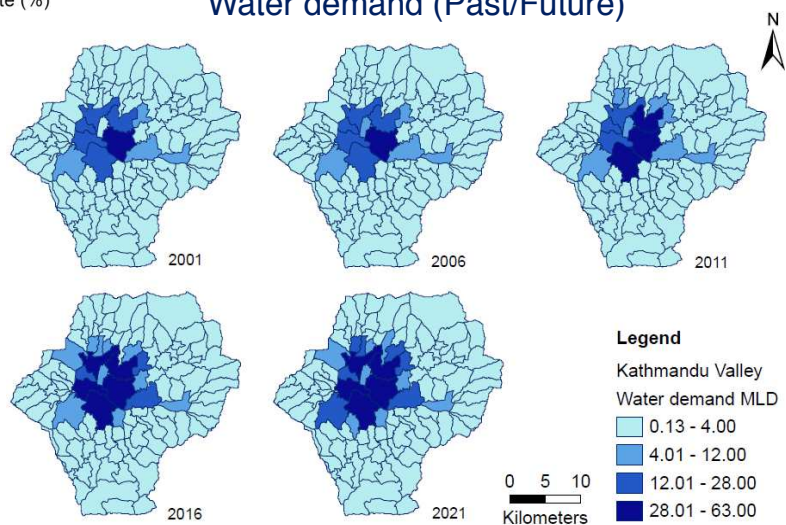
P_t – Population at time t

P_0 – Population at time t_0

r – Growth rate

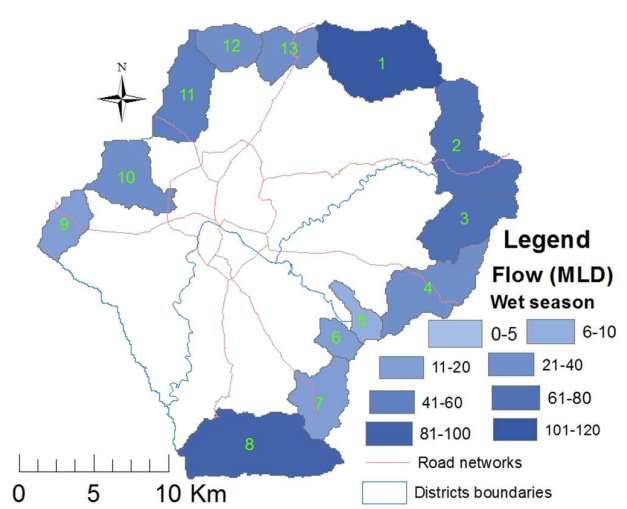
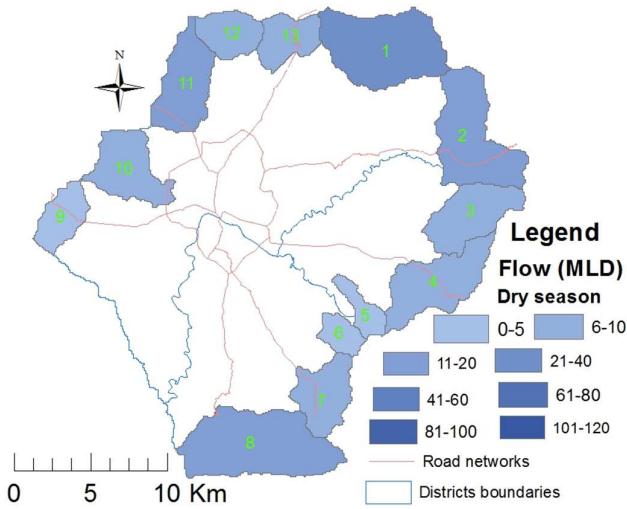
t – Time (number of periods)

Water demand (Past/Future)



Water Security Map (WG1)

Surface Water Resources in Mountainous region

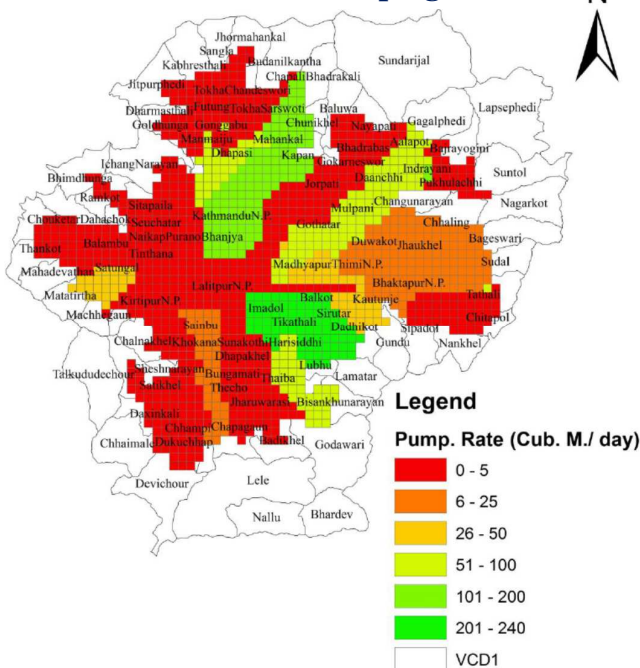


Thapa, B.R., Ishidaira, H., Bui, T.H. and Shakya, N.M. (2016):
 Evaluation of water resources in mountainous region of Kathmandu Valley using high resolution satellite precipitation product,
Journal of Japan Society of Civil Engineering, Ser. G (Environment), Vol.72(5), 1_27-1_33; doi:10.2208/jscej.72.1_27

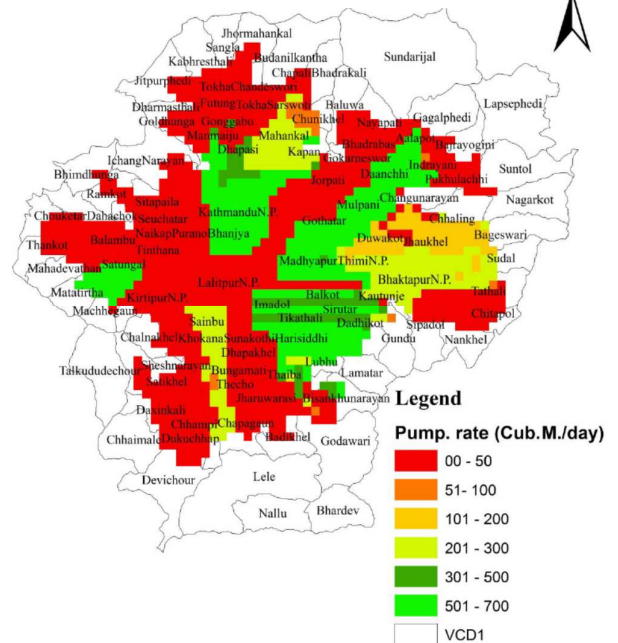
Water Security Map (WG1)

Shallow Groundwater

VDCwise 2016 Pumping Rate



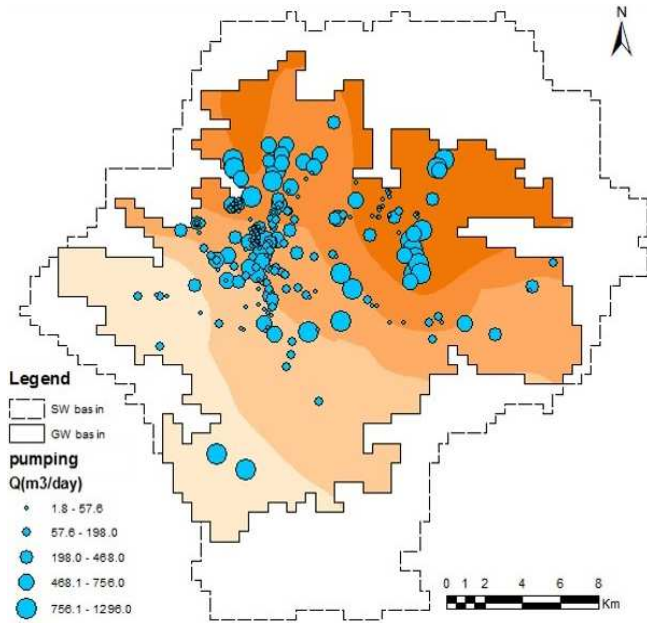
VDCwise Proposed Max. Pumping Rate



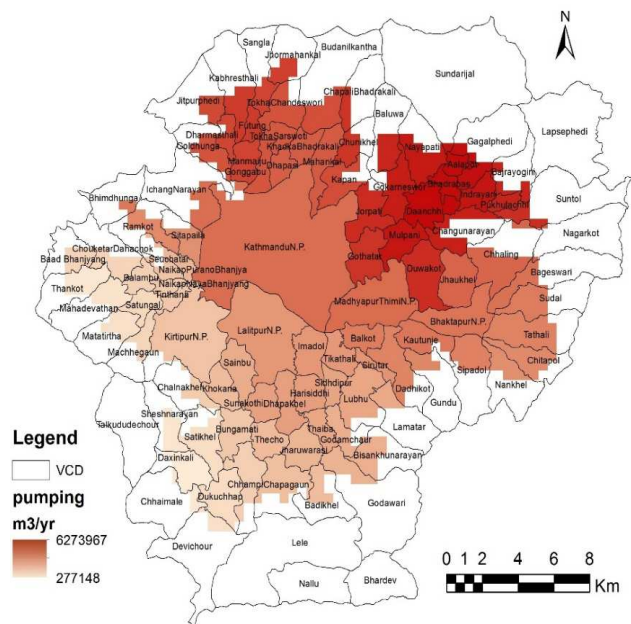
Water Security Map (WG1)

Deep Groundwater

Pumping (1999) comparison with proposed extraction rate

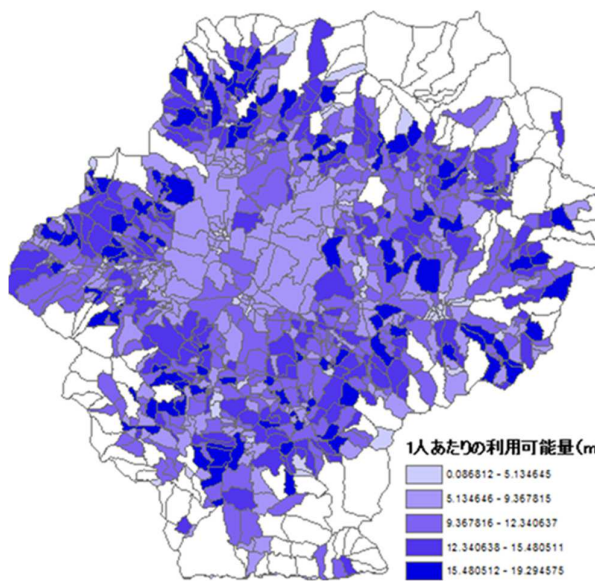


VDC wise proposed extraction rates

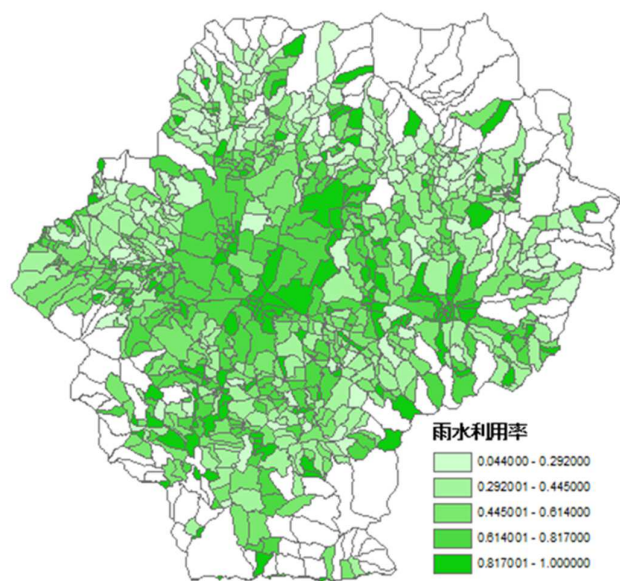


Water Security Map (WG1)

Potential of Rain Water Harvesting (Roof-top)



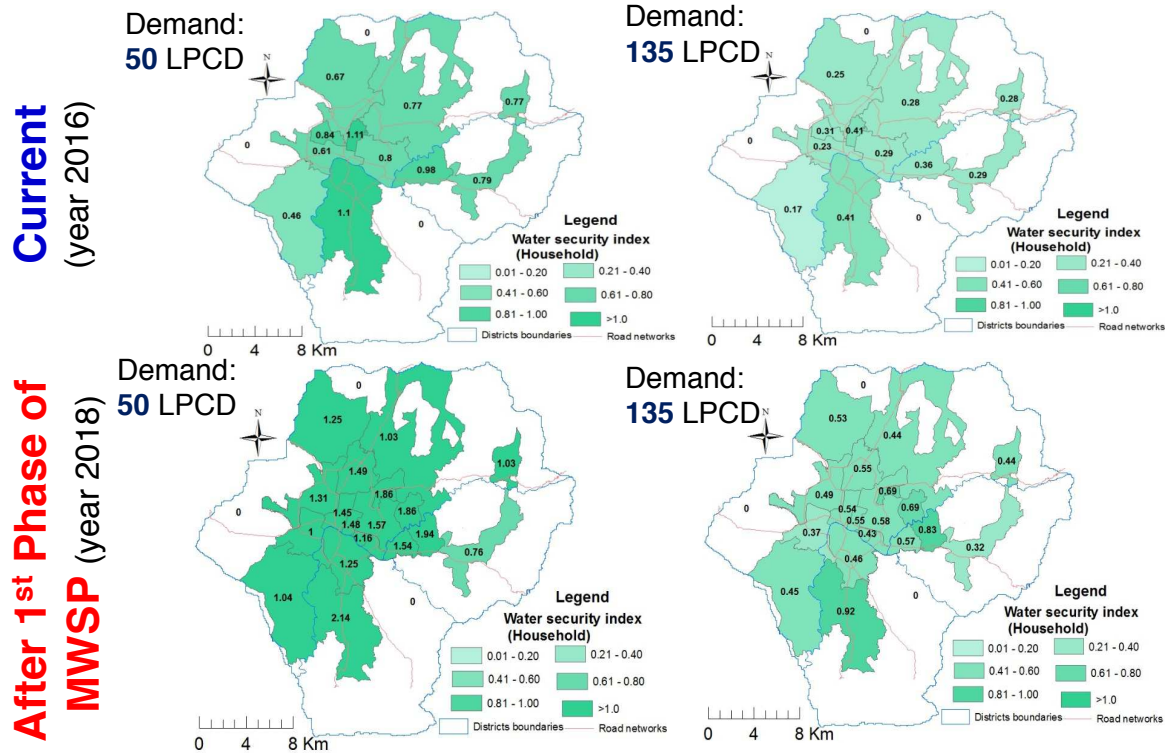
RWH potential [m³/y/captia]



Fraction of Rainwater Use

Water Security Map (WG1)

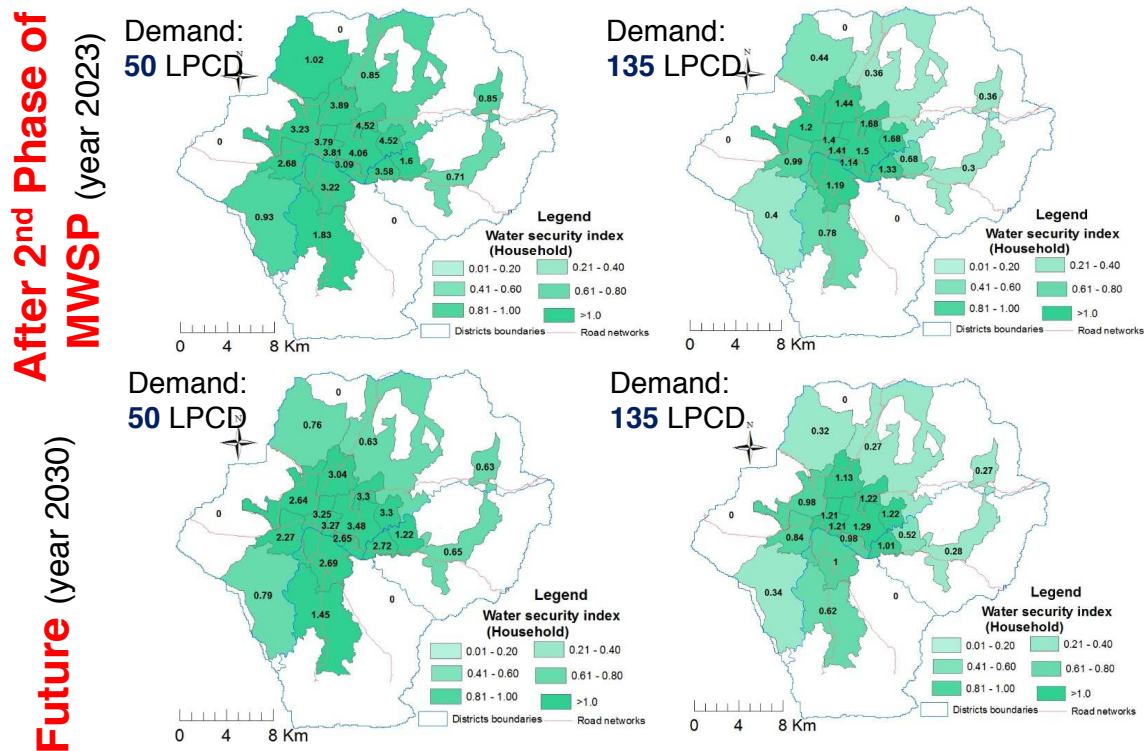
Spatial Distribution of Household Water Security Index (WSI)



Thapa B.R., Ishidaira H., Pandey V.P., Bhandari T.B. and Shakya N.M. (2018): Evaluation of Water Security in Kathmandu Valley before and after Water Transfer from another Basin, *Water* 2018, 10(2), 224; <https://doi.org/10.3390/w10020224>

Water Security Map (WG1)

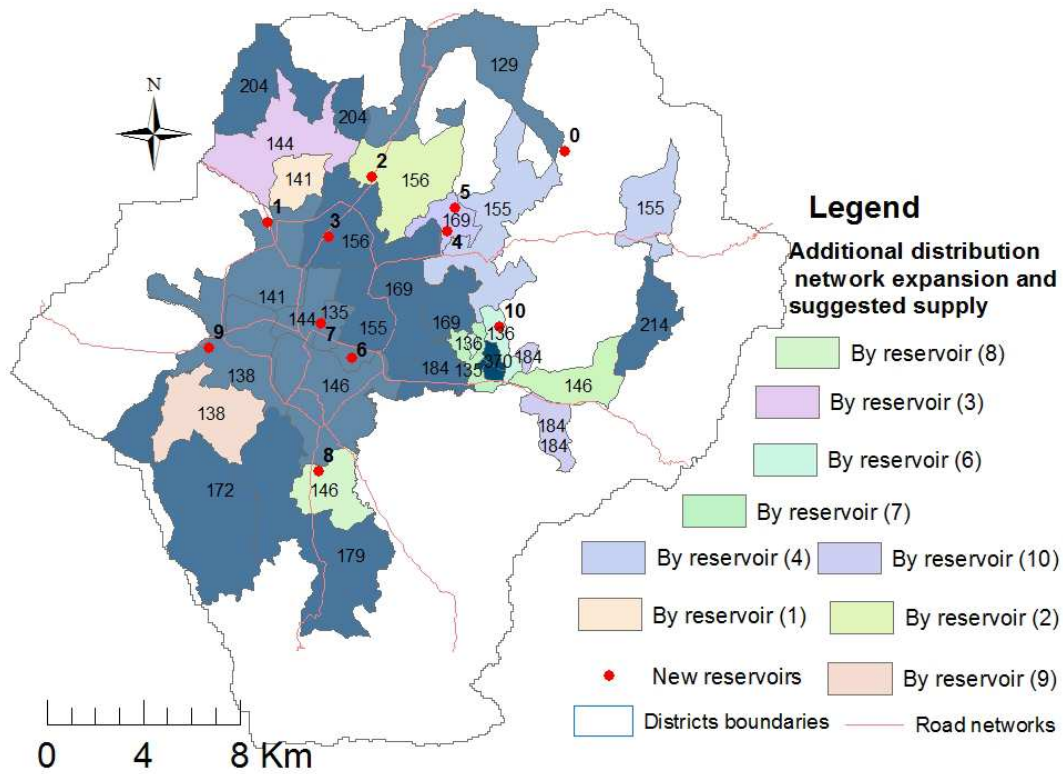
Spatial Distribution of Household Water Security Index (WSI)



Thapa B.R., Ishidaira H., Pandey V.P., Bhandari T.B. and Shakya N.M. (2018): Evaluation of Water Security in Kathmandu Valley before and after Water Transfer from another Basin, *Water* 2018, 10(2), 224; <https://doi.org/10.3390/w10020224>

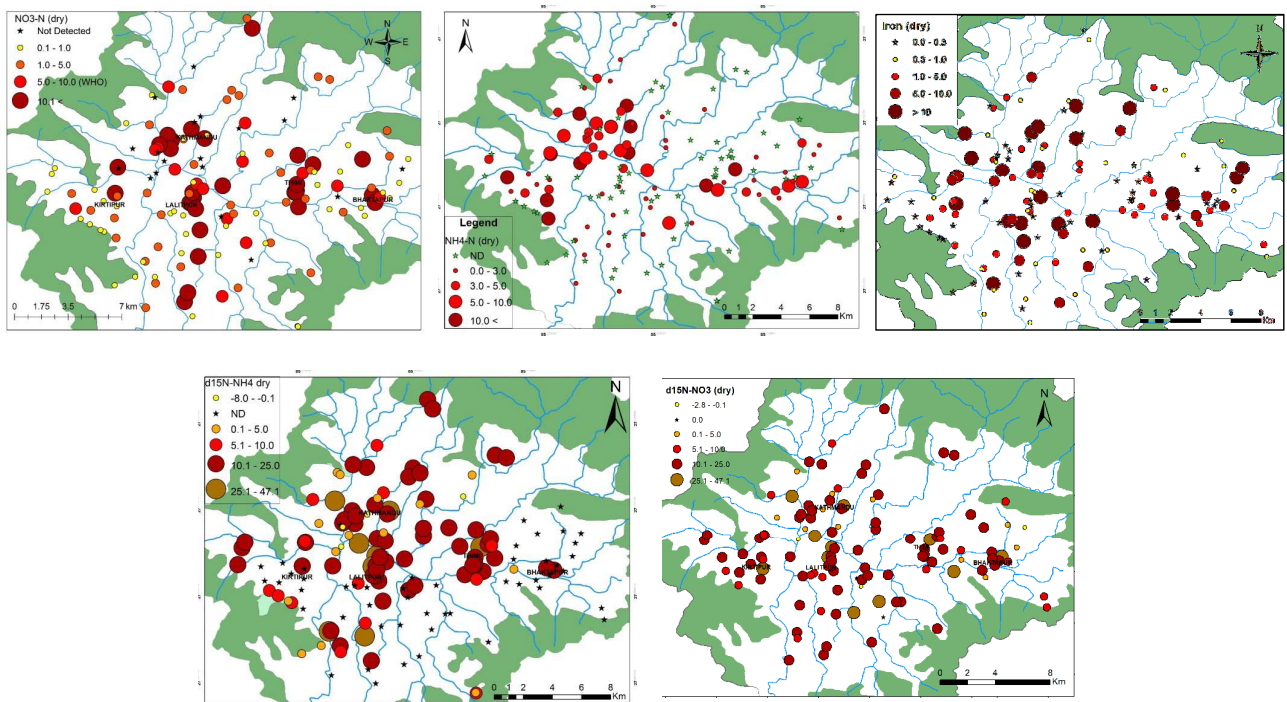
Water Security Map (WG1)

Strategies for Reallocation of potable water after completion of MWSP for equitable distribution of water



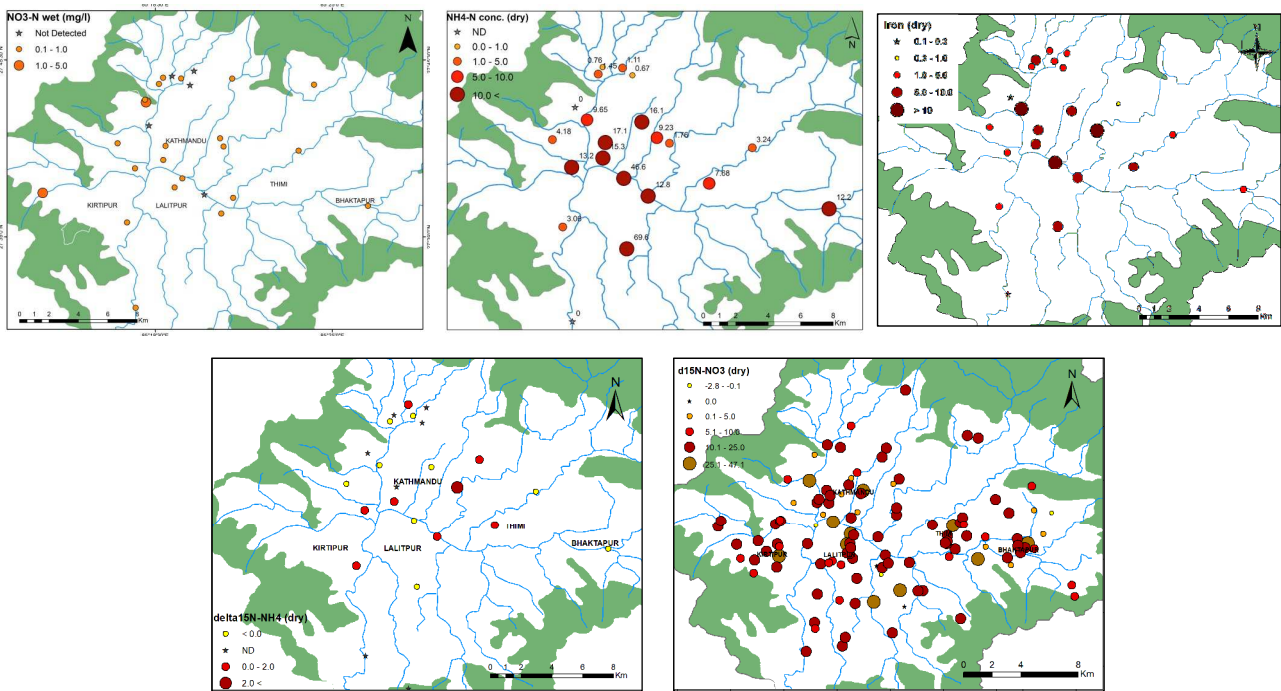
Water Security Map (WG2)

Shallow well water quality and isotope values in the dry season (2016)



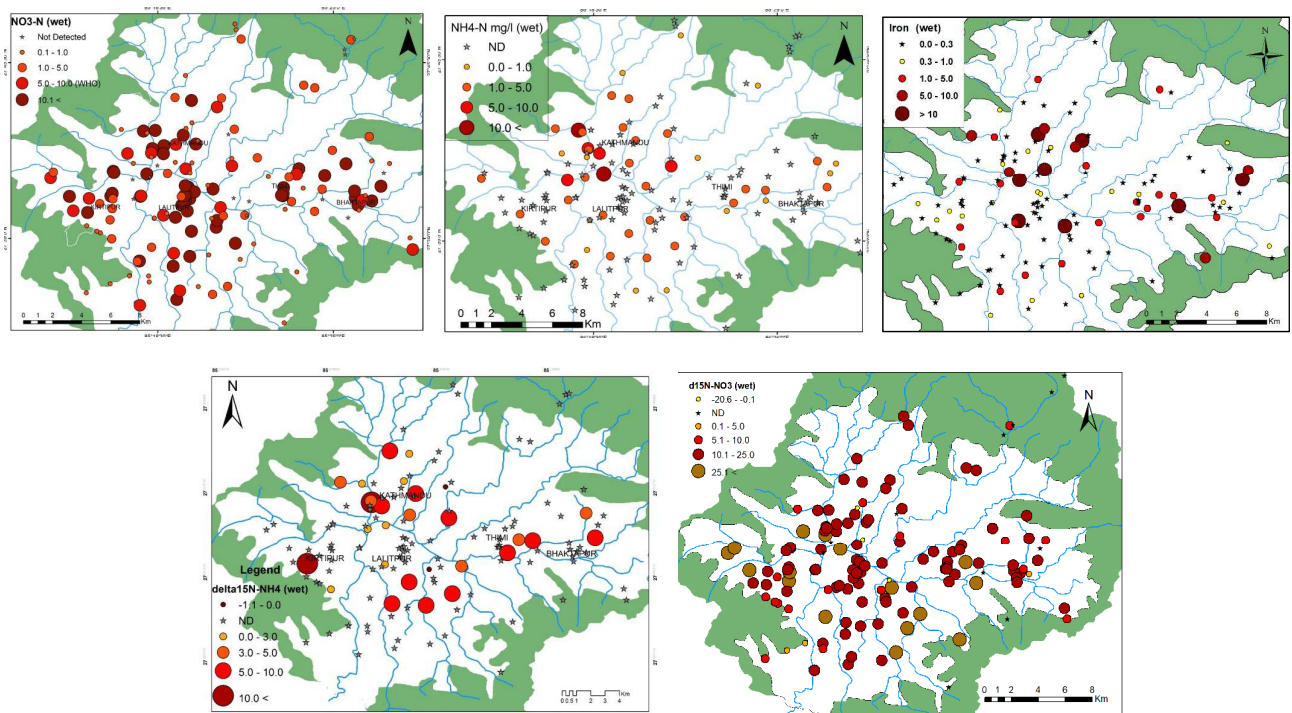
Water Security Map (WG2)

Deep well water quality and isotope values in the dry season (2016)



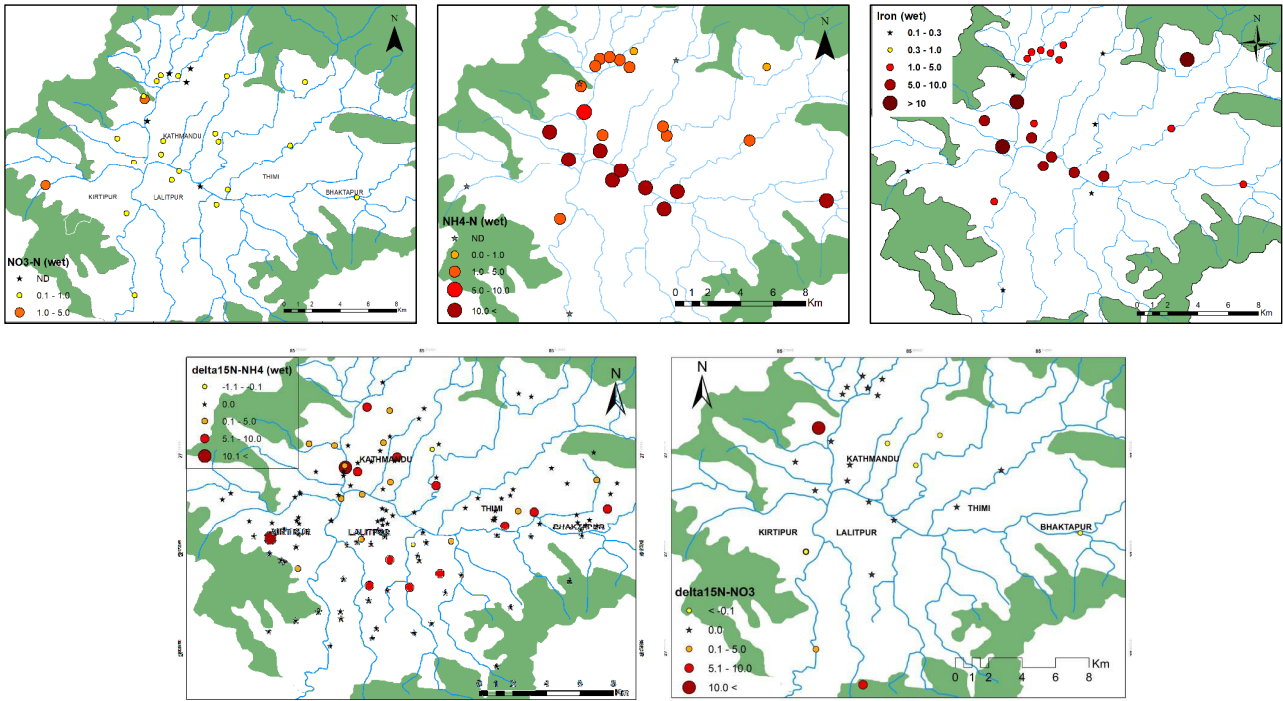
Water Security Map (WG2)

Shallow well water quality and isotope values in the wet season (2016)



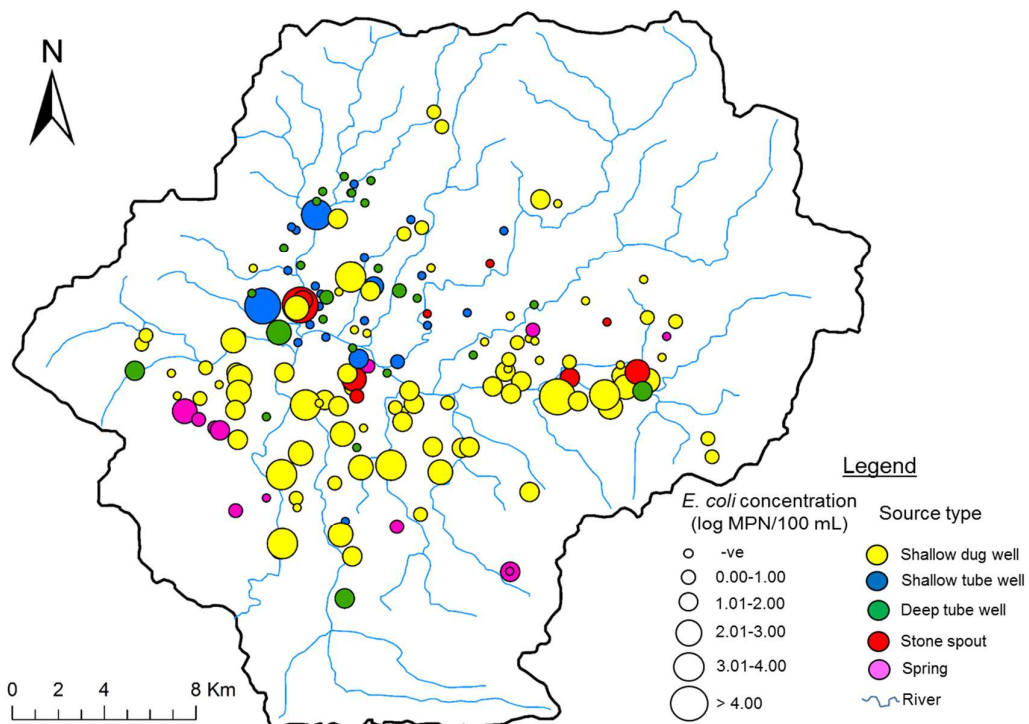
Water Security Map (WG2)

Deep well water quality and isotope values in the wet season (2016)



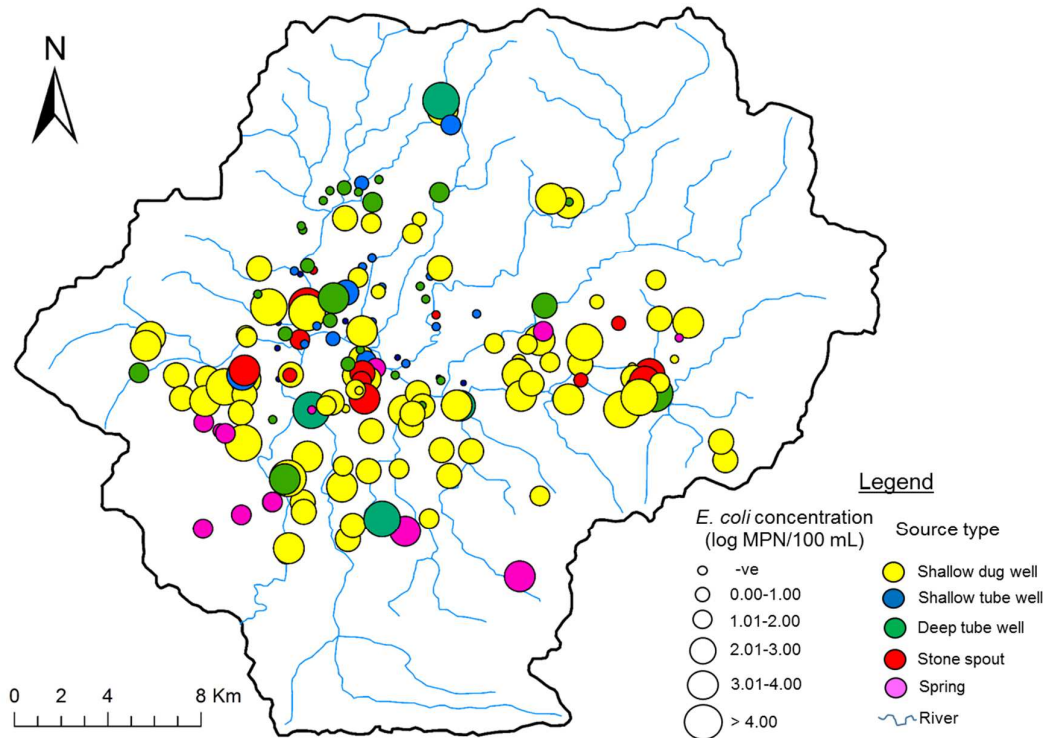
Water Security Map (WG3)

E. coli concentration in the dry season (2016)



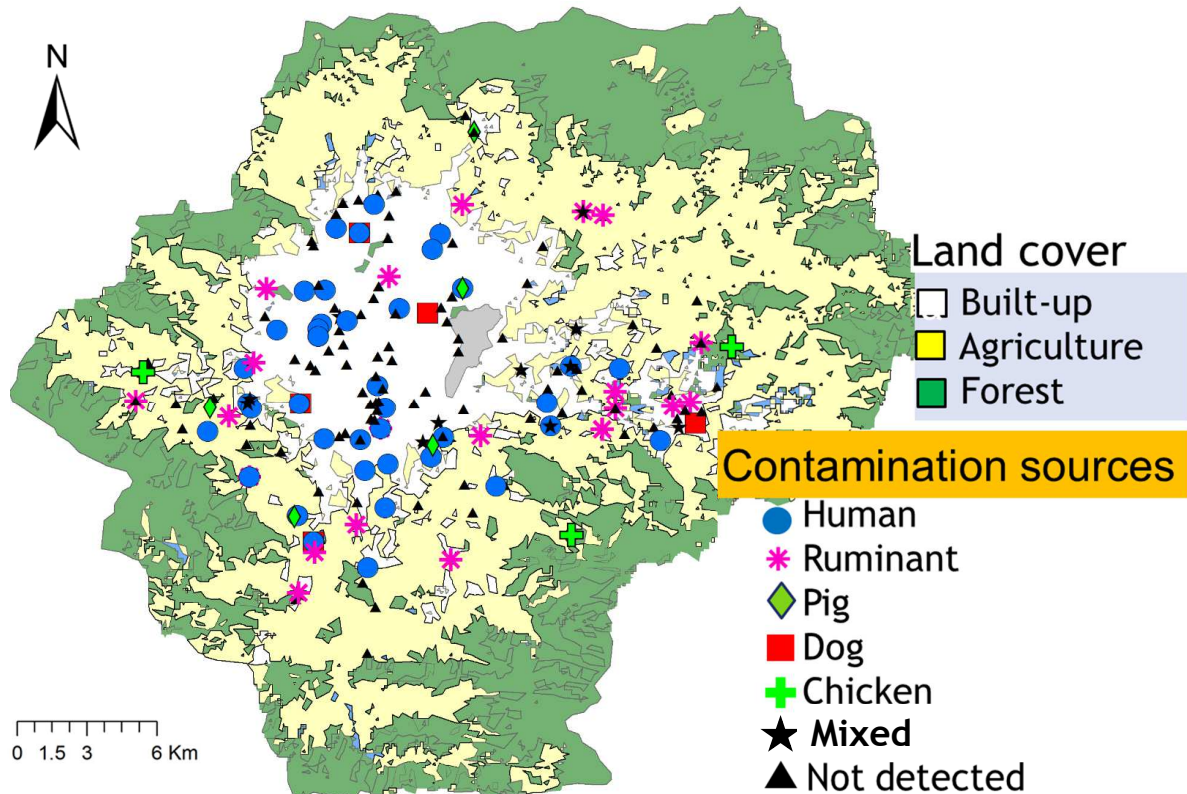
Water Security Map (WG3)

E. coli concentration in the wet season (2016)



Water Security Map (WG3)

Sources of fecal contamination in groundwater samples (2016)



Water Security Map (WG5)

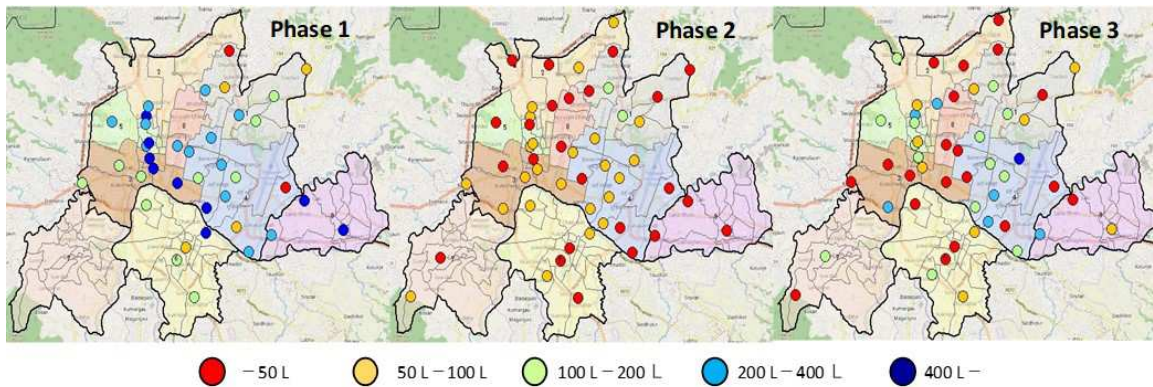
Household Survey

Face to face questionnaire survey for 1500 households: 30 HHs in each of 50 clusters (locations) selected randomly based on probability populational to HH size of each ward

Period of questionnaire survey

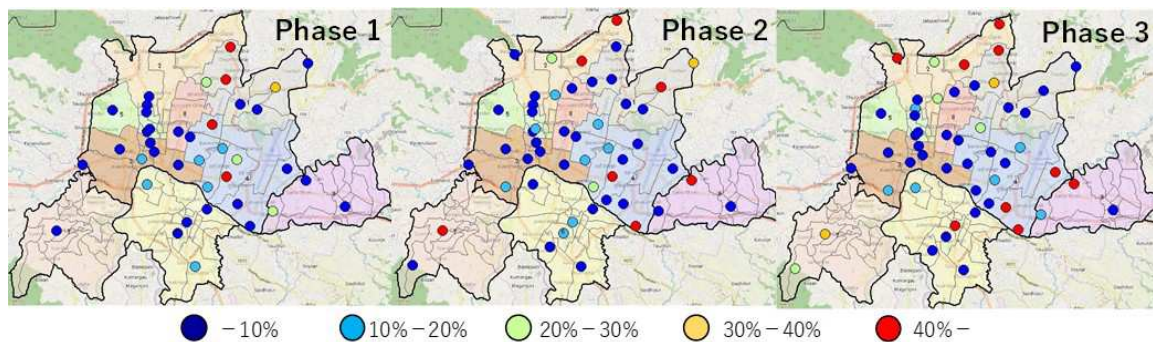
- Phase 1: Jan. – April, 2015 (Dry season, Before GE)
- Phase 2: Dec., 2015 – April, 2016 (Dry season, After GE)
- Phase 3: Aug. – Nov., 2016 (Wet season)

Household Survey: Pipe Water Supply Hours per Week

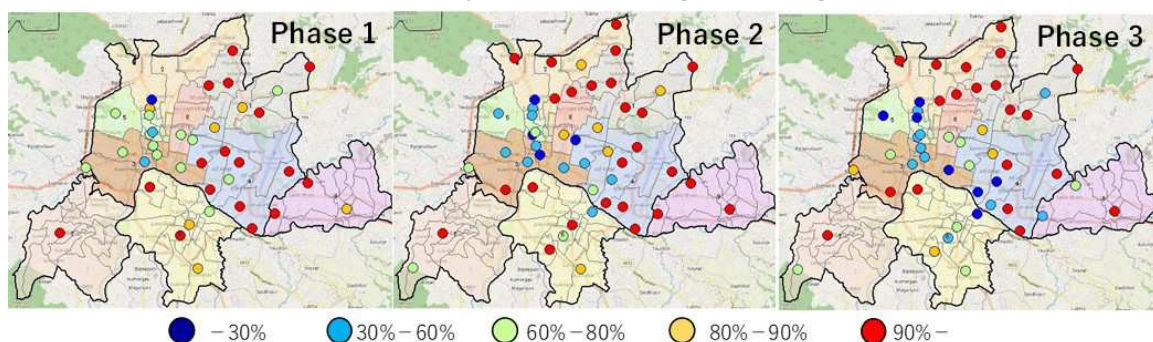


Water Security Map (WG5)

Household Survey : HHs using groundwater for drinking and cooking

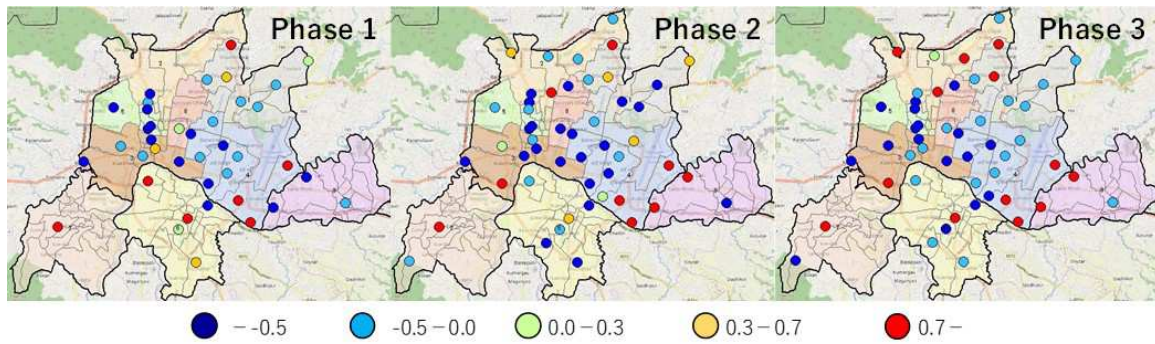


Household Survey: HHs Treating Drinking Water

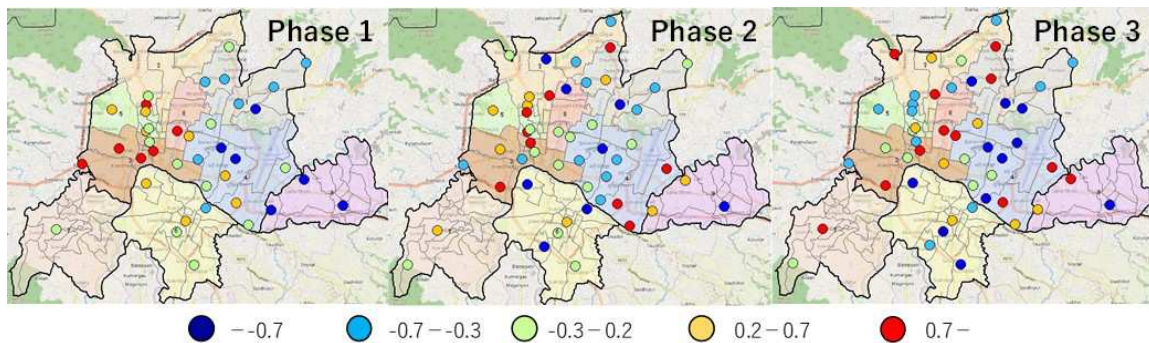


Water Security Map (WG5)

Household Survey: Water Stress Index (Inconvenience in Daily Life)

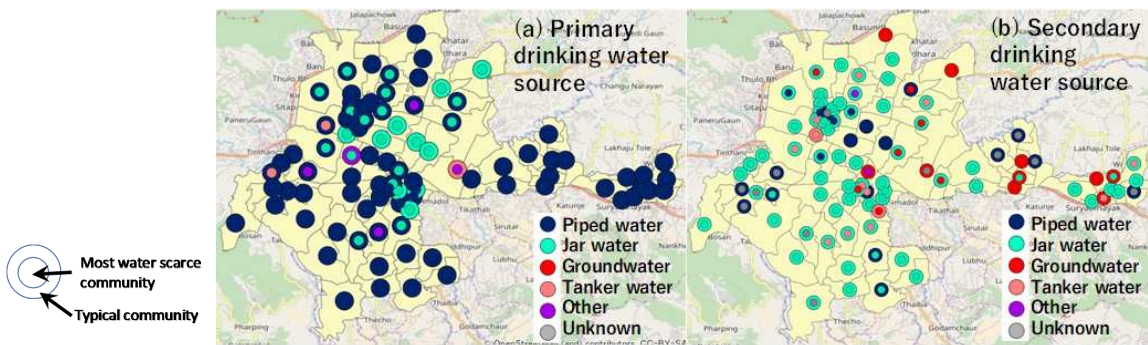


Household Survey : Water Stress Index (Anxiety to Water Safety)

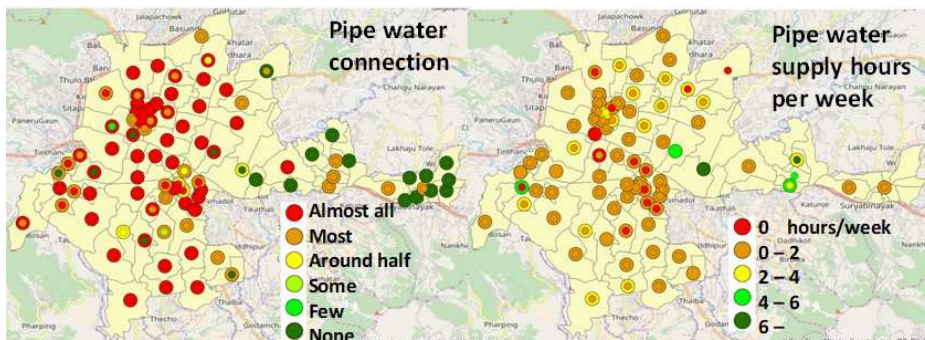


Water Security Map (WG5)

Community Survey: Main Source of Drinking Water

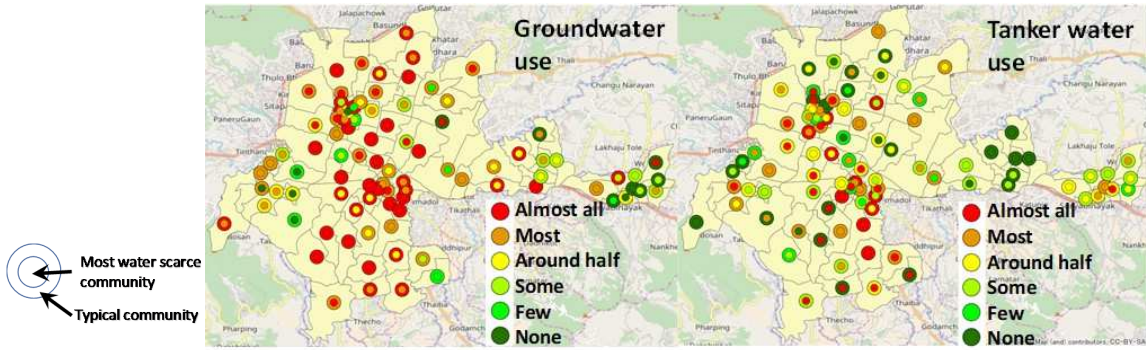


Community Survey: Pipe Water Connection and Supply Hours

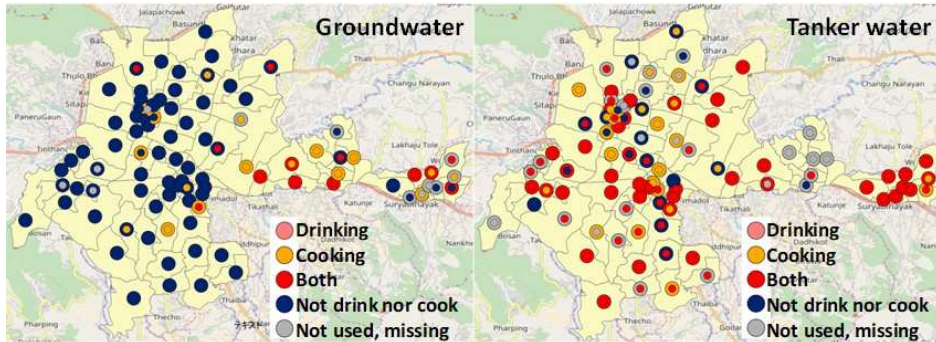


Water Security Map (WG5)

Community Survey: Alternative Water Use

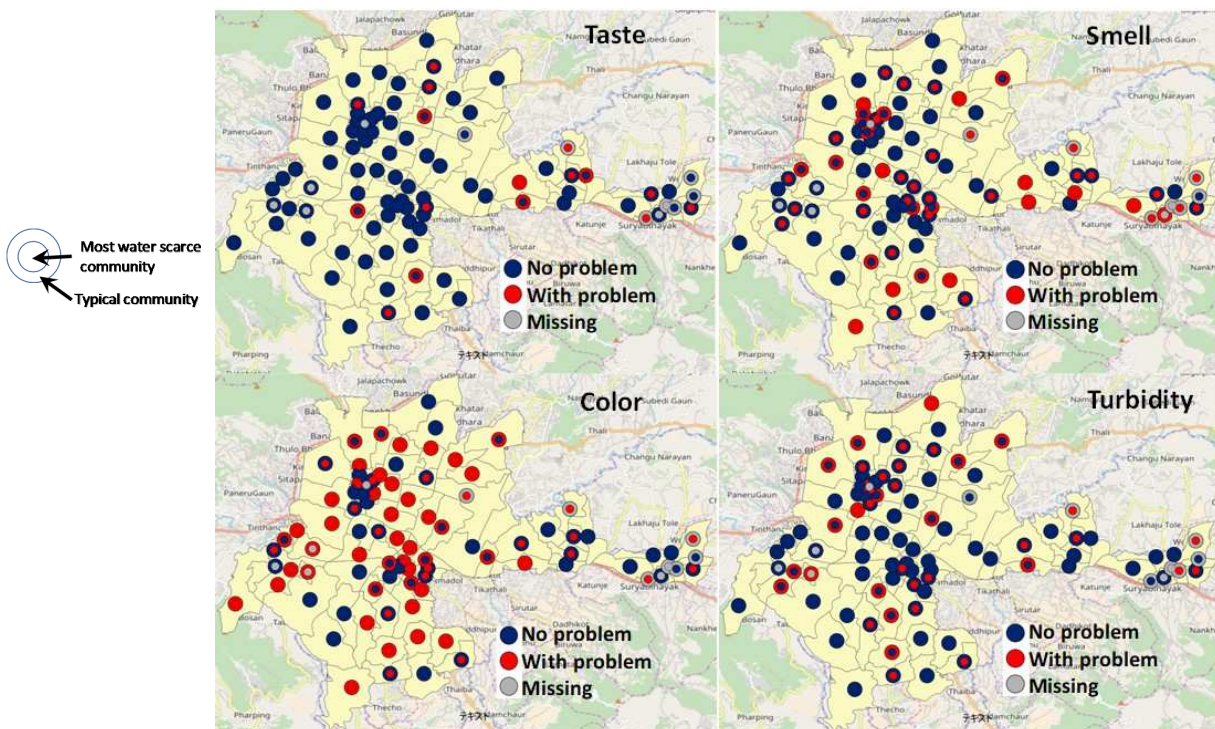


Community Survey: Alternative Water Use for Drinking and Cooking



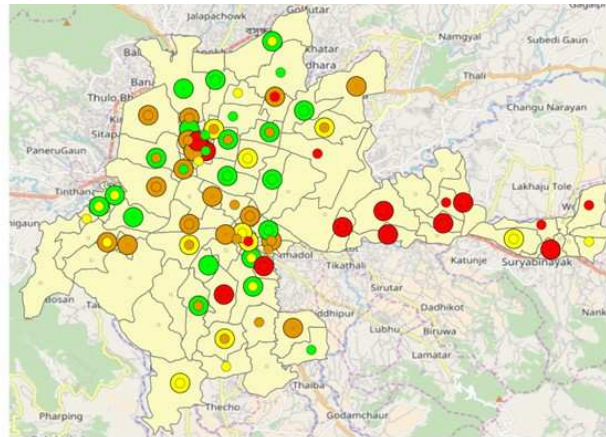
Water Security Map (WG5)

Community Survey: Groundwater Quality



Water Security Map (WG5)

Community Survey: Candidate Communities for LCD System Installation in Priority Based on Groundwater Quality and Ingested Water

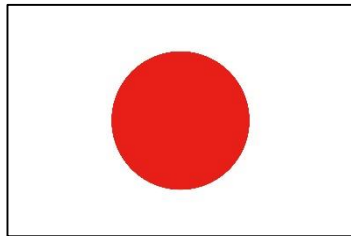


For all communities indicated, groundwater quality is poor

- Ingesting groundwater
- Ingesting tanker water with poor quality
- Ingesting tanker water
- Ingesting Jar water

LCD* system for sustainable safe water distribution

*LCD : Locally fitted, compact, and distributed



57



Sustainable safe drinking water distribution in developing country by LCD system

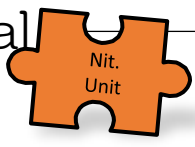
Sustainable safe water supply is urgently needed in developing countries. The LCD system is composed of locally available materials and is used as a contaminant removal technology suitable for the local economic level. LCD application makes sustainable safe water supply possible. Here, we propose LCD units for the removal of major contaminants: Iron, Ammonia, and Nitrate.

*LCD : Locally fitted, compact, and distributed

① Iron removal
Easy maintenance
Sponge filtration unit




② Ammonia removal
No aeration energy requirement
Nitrification unit




③ Nitrate removal
Easy application
Denitrification unit




Combine systems to meet people demand

Case 1: Domestic water use



Iron stains my white dress!

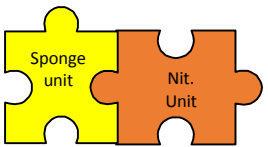
Target : Iron
System: Sponge filtration



My water smell like sewage!



Target : Iron, Ammonia
System : Sponge + Nitrification



Case 2: Drinking water use



My water exceed quality standard!

Target : Nitrate
System : Denitrification unit



No way to use contaminated water!



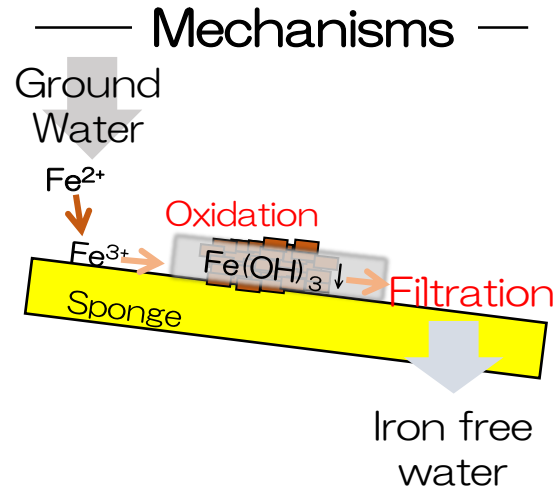
Target : Iron, Ammonia, Nitrate
System : Sponge+Nitrification+Denitrification units



Sponge Filtration Unit

Information

Targets : Iron, Turbidity removals
Water uses: Bathing, Washing
Capacity: 1,000-4,000 L in a day
Fe₂⁺ Removal: >99%
Turbidity removal
(54 → 0 NTU)



System Types

- Community water supply (Cap: <4,000L/day)



- House hold water supply (Cap: < 1,000L/day)



Nitrification unit



Information

Targets : Ammonia removal
Water uses: Bathing, Drinking
Capacity: 1,000 – 3,000 L/day
NH₄⁺ Removal: >99%

Reference

Khanitachalcha, W., Shakya, M., Nakano, Y., Tanaka, Y., and Kazama, F. (2012) Development of an attached growth reactor for NH₄⁺-N removal at a drinking water supply system in Kathmandu Valley, Nepal. Journal of Environmental Science and Health, Part A, 47(5).

System Type

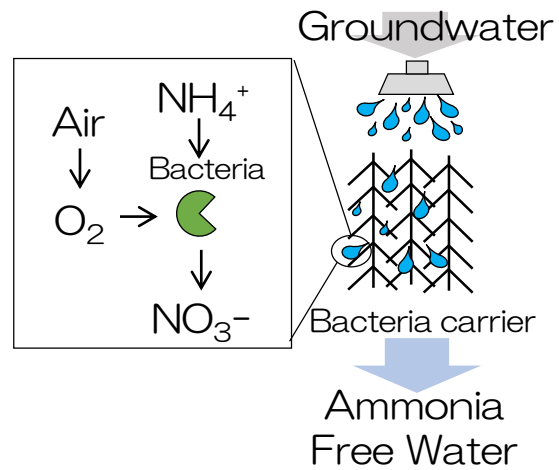
- Community water supply (Cap: <3,000L/day)



- House hold water supply (Cap: < 1,000L/day)



Mechanisms



Denitrification Unit



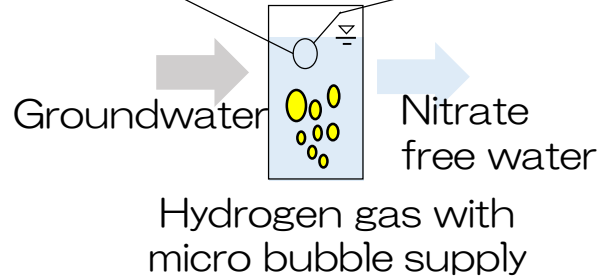
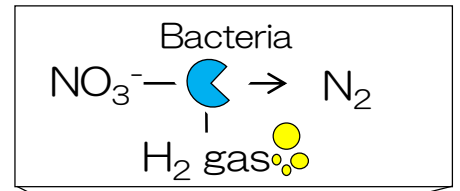
Information

Target : Nitrate Removal
Water use : Drinking
Capacity : 25-500 L/day
NO₃⁻ Removal: > 97%

Reference

Eamrat, R., Mochizuki, T., Kamei, T., and Kazama, F. (2016) Hydrogenotrophic Denitrification Activity under Intermittent Hydrogen Supply using Micro-Bubble System-hydrogenotrophic Denitrification Activity under Intermittent Hydrogen Supply using Micro-Bubble System. Naresuan University Engineering Journal, 11, 47-51. [online]

— Mechanism —



System Type

- Community water supply (Cap.: < 500L/day)



- House hold water supply (Cap.: < 25L/day)



Analytical Methods

for Water sample

Target : N, P , Metal, COD
and HCO_3^-

Ver. 3

Oct.25, 2016



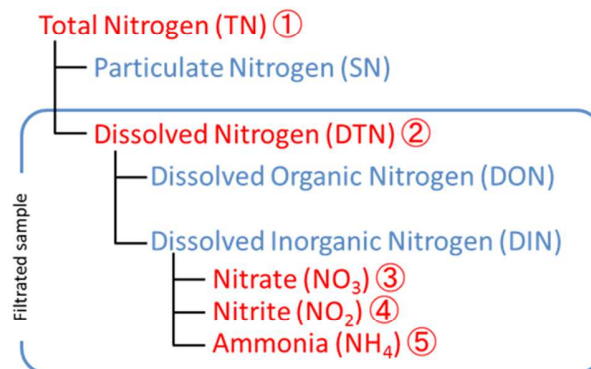
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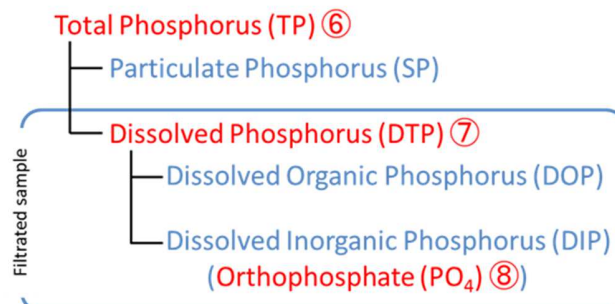
List of Lecture on analysis

①—⑬ : analysis targets

1. Nitrogen



2. Phosphorus



3. Metal

- Total Iron (T-Fe) ⑨
- Acid soluble Iron (AS-Fe) ⑩
include: Fe²⁺, Fe³⁺, Iron complex compound, Fe(OH)₃, Clay particles...
These can change easily a form by an environmental change.
- Ferrous ion (Fe²⁺) ⑪
- Dissolved Manganese (D-Mn) ⑫

4. Chemical Oxygen Demand (COD_{Cr}) ⑬

5. Bicarbonate (HCO₃⁻) ⑭

▪ Method for Cleaning Equipment

- before use in experiment (for volumetric flask)

Rinse the volumetric flask in pure water 3 times

- cleaning solution

Put cleaning powder (50g), and pour water (10L) into the bucket.

* the ultrasonic cleaning machine: 30g cleaning powder/10L

This solution can be use several times.

- after use in experiment

1. Throw away the inside (if there are stains, rub equipments with a sponge).

2. Wash equipments in (tap) water several times.

3. Fill equipment with the cleaning solution of the bucket, sink equipments, and keep overnight.

* if you use the ultrasonic cleaning machine, set a timer at 20 minutes, and start to wash.

4. Take out equipments.

5. Wash equipments with tap water until a bubble disappears.

6. Rinse equipments in pure water 3 times.

7. Upend equipments to the basket, and put it in a drying cabinet.

8. After dry, cap the bottle, or put the equipments into a bag, then keep in shelf.

▪ Sampling and storage method of sample

- Target : nonmetal

《sampling》 Bring back samples to the laboratory. If it need to filter, filtration do in laboratory.

《storage》 Stored in refrigerator (analysis period is less than 1week),
Keep in freezer (more than 1week)

- Target : metal

《sampling》 Fill bottle directly from sampling source and stopper (if it need to filter, filtration do onsite).

Sample with 2ml-HCl/100ml-sample at time of collection.

《storage》 Stored at room

▪ **Preparation of Standard Solution (Weight method)**

Equipment :

- pipette (10mL, 5mL, 1mL)
- Screw Bottle (30mL)
- Pure water

Apparatus :

- chemical balance

Procedure :

1. Power on chemical balance. Wait until end of calibration.
2. Put a Screw bottle on the balance, close the cover, wait, and push a "tare" button.
3. Weigh out the required amount of "Stock sol." or "Std. Sol. A (5mg/L)" (refer to table 1-6), and close a cover.
4. Wait, then record actual weight.
5. Add pure water until total weight to 25 gm.
6. Wait, then record actual gross weight.
7. Cap a Screw bottle, and mix it well.
8. Calculation of concentration (mg/L)

$$\text{Actual concentration} = \text{concentration of Std. Sol. (or A)} \times \frac{\text{actual weight(g)}}{\text{actual gross weight(g)}}$$

①-③ TN/DTN/NO₃-N Standard solutions: (0.05 ~ 5 mg-N/L)

Chemical : Nitrate Nitrogen Standard Solution (1000*mg- NO₃-N/l)

*when calculate, use the written value of the standard solution bottle.

Table.1 TN/DTN/NO₃-N Standard solutions

Required concentration : mg-N/L	5 (Std. Sol. A)	4	3	2	1	0.5	0.2	0.1	0.05
weight of Std. sol. (1g/L) : g	0.125	0.10	0.075	0.050	—	—	—	—	—
weight of Std. sol. A (5mg/L) : g	—	—	—	—	5.0	2.5	1.0	0.5	0.25
actual weight : g									
actual gross weight : g									
Actual concentration : mg-N/L									

③-2 NO₃-N Standard solutions of second derivative method: (5 ~ 25 mg-N/L)

Chemical : Nitrate Nitrogen Standard Solution (1000*mg- NO₃-N/l)

*when calculate, use the written value of the standard solution bottle.

Table.2 NO₃-N Standard solutions of second derivative

Required concentration : mg-N/L	25	20	15	10	5
weight of Std. sol. (1g/L) : g	0.625	0.50	0.375	0.25	0.125
actual weight : g					
actual gross weight : g					
Actual concentration : mg-N/L					

④ NO₂-N Standard solutions: (0.02 ~ 0.5 mg-N/L)

Chemical : Nitrite Nitrogen Standard Solution (1000*mg- NO₃-N/l)

*when calculate, use the written value of the standard solution bottle.

Table.3 NO₂-N Standard solutions

Required concentration : mg-N/L	5 (Std. Sol. A)	0.5	0.2	0.1	0.05	0.02
weight of Std. sol. (1g/L) : g	0.125	—	—	—	—	—
weight of Std. sol. A (5mg/L) : g	—	2.5	1.0	0.5	0.25	0.1
actual weight : g						
actual gross weight : g						
Actual concentration : mg-N/L						

⑤ NH₄-N Standard solutions: (0.05 ~ 5 mg-N/L)

Chemical : Ammonium Nitrogen Standard Solution (1000*mg- NO₃-N/l)

*when calculate, use the written value of the standard solution bottle.

Table.4 NH₄-N Standard solutions

Required concentration : mg-N/L	5 (Std. Sol. A)	4	3	2	1	0.5	0.2	0.1	0.05
weight of Std. sol. (1g/L) : g	0.125	0.10	0.075	0.050	—	—	—	—	—
weight of Std. sol. A (5mg/L) : g	—	—	—	—	5.0	2.5	1.0	0.5	0.25
actual weight : g									
actual gross weight : g									
Actual concentration : mg-N/L									

⑥-⑧ TP/DTP/PO₄-P Standard solutions: (0.01 ~ 5 mg-P/L)

Chemical : Phosphorus Standard Solution (1000*mg- PO₄-P/l)

*when calculate, use the written value of the standard solution bottle.

Table.5 TP/DTP/PO₄-P Standard solutions

Required concentration : mg-P/L	5 (Std. Sol. A)	4	3	2	1	0.5	0.2	0.1	0.05
weight of Std. sol. (1g/L) : g	0.125	0.10	0.075	0.050	—	—	—	—	—
weight of Std. sol. A (5mg/L) : g	—	—	—	—	5.0	2.5	1.0	0.5	0.25
actual weight : g									
actual gross weight : g									
Actual concentration : mg-P/L									

⑨-⑪ T-Fe / AS-Fe/ Fe²⁺ Standard solutions: (0.2 ~ 1 mg/L)

Chemical : Iron Standard Solution (1000*mg/l)

*when calculate, use the written value of the standard solution bottle.

**need to add conc. HCl at all bottles. Because iron absorb to bottle.

Table.6 Fe²⁺/T-Fe Standard solutions

Required concentration : mg-Fe/L	5 (Std. Sol. A)	1	0.8	0.6	0.4	0.2
weight of Std. sol. (1g/L) : g	0.125	—	—	—	—	—
weight of Std. sol. A (5mg/L) : g	—	5.0	4.0	3.0	2.0	1.0
actual weight : g						
Add conc .HCl (ml)	0.3	0.3	0.3	0.3	0.3	0.3
actual gross weight : g						
Actual concentration : mg-Fe/L						

⑫ D-Mn Standard solutions: (0.1 ~ 4 mg/L)

Chemical : Manganese Standard Solution (1000*mg/l)

*when calculate, use the written value of the standard solution bottle.

Table.7 T-Mn Standard solutions

Required concentration : mg-Mn/L	5 (Std. Sol. A)	4	3	2	1	0.5	0.1
weight of Std. sol. (1g/L) : g	0.125	0.10	0.075	0.050	—	—	—
weight of Std. sol. A (5mg/L) : g	—	—	—	—	5.0	2.5	0.5
actual weight : g							
actual gross weight : g							
Actual concentration : mg-Mn/L							

① Total Nitrogen Analysis by Ultraviolet Spectrophotometric Screening Method

(ref. : Standard Methods for the Examination of Tap Water in Japan)

***volume is different**

Equipment:

- 200ml measuring cylinder
- transfer pipet
- pipette(10ml, 1ml)
- bottle
- 100 ml Beaker
- tubes with cap
- silica cuvette(1cm) : ***cannot use glass cuvette, that block UV light.**

Apparatus:

- Analytical balance
- Spectrophotometer

Required Chemicals:

1. Distilled water
2. Potassium persulfate (freshly prepared) : for analysis of Nitrogen
3. Sodium hydroxide : for analysis of Nitrogen
4. Conc. HCl : G.R.
5. Nitrate Nitrogen Standard Solution (1000mg- NO₃-N/l)

Preparation of reagents:

1. Sodium hydroxide/Potassium persulfate (Digestion Solution): (0.4ml/tube)
***Total Nitrogen are converted to Nitrate.**
Dissolve 4gm NaOH to 100 ml distilled water, stir it with the help of magnetic stirrer.
After dissolve, add 3 gm of Potassium persulfate. Then stir.
2. Hydrochloric acid solution, HCl, 1N : same of Nitrate (0.48ml/tube)
To 110ml of distilled water, add 10 ml of Hydrochloric Acid
3. Preparation of Nitrate Standard solutions: (0.05 ~ 5 mg/l) same of Nitrate
See Table.1

Procedure: see Fig. 1

a. **Sample Preparation:**

- i. triplicate all the samples
- ii. Take 2ml of samples.
(Dilute the sample (if required) and take 2 ml of each)
* had better check nitrate concentration by pack test.
- iii. Add 0.4 ml of Digestion Solution to each sample.
- iv. Autoclave all the samples for 30 minutes at 121°C.
- v. After cooling, add 0.48ml of the 1N HCl to the samples, and mix.
- vi. Wait until hydroxide precipitates.

b. **Standard solutions preparation:**

- duplicate blank and all the standard solutions
Treat blank and std. solutions in same manner as samples.

c. **Photometric measurement:**

- Measure the absorbance at 220nm and 275nm.
220nm : to obtain NO₃⁻ reading
275nm : to determine interference due to dissolved organic matter

Calculation:

For samples, blank and std. solutions, subtract two times the absorbance reading at 275nm from the reading at 220nm to obtain absorbance due to NO₃⁻:

$$2DS = Abs_{220} - 2 * Abs_{275}$$

where:

- 2DS = absorbance due to NO₃⁻
Abs₂₂₀ = the absorbance reading at 220nm
Abs₂₇₅ = the absorbance reading at 275nm.

Construct a standard curve by plotting absorbance due to NO₃⁻(2DS) against NO₃⁻-N concentration of standard. Using corrected sample absorbances, obtain sample concentrations directly from standard curve.

② Dissolved Nitrogen Analysis by Ultraviolet Spectrophotometric Screening Method

(ref. : Standard Methods for the Examination of Tap Water in Japan)

***volume is different**

Equipment:

- 200ml measuring cylinder
- transfer pipet
- pipette(10ml, 1ml)
- bottle
- 100 ml Beaker
- tubes with cap
- silica cuvette(1cm) : ***cannot use glass cuvette, that block UV light.**
- syringe
- syringe filter (0.2 μ m) : i.e. cellulose acetate

Apparatus:

- Analytical balance
- Spectrophotometer

Required Chemicals:

1. Distilled water
2. 3 gm, Potassium persulfate (freshly prepared) : for analysis of Nitrogen
3. 4 gm, Sodium hydroxide : for analysis of Nitrogen
4. 10 ml, Conc. HCl : G.R.
5. Nitrate Nitrogen Standard Solution (1000mg- NO₃-N/l)

Preparation of reagents:

1. Sodium hydroxide/Potassium persulfate (Digestion Solution): (0.4ml/tube)

***Dissolved Nitrogen are converted to Nitrate.**

Add 4gm NaOH to 100 ml distilled water, stir it with the help of magnetic stirrer. After dissolve, add 3 gm of Potassium persulfate. Then stir.

2. Hydrochloric acid solution, HCl, 1N : same of Nitrate (0.48ml/tube)

To 110ml of distilled water, add 10 ml of Hydrochloric Acid

3. Preparation of Nitrate Standard solutions: (0.05 ~ 5 mg/l) same of Nitrate

See Table.1

Procedure: see Fig. 1

- a. Removal of suspended particles:
Filter through a syringe filter (0.2µm)
- b. Sample Preparation:
 - i. duplicate all the samples
 - ii. Take 2ml of samples.
(Dilute the sample (if required) and take 2 ml of each)
* had better check nitrate concentration by pack test.
 - iii. Add 0.4 ml of Digestion Solution to each sample.
 - iv. Autoclave all the samples for 30 minutes at 121°C.
 - v. After cooling, add 0.48ml of the 1N HCl to the samples, and mix.
 - vi. Wait until hydroxide precipitates.
- c. Standard solutions preparation:
duplicate blank and all the standard solutions
Treat blank and std. solutions in same manner as samples.
- d. Photometric measurement:
Measure the absorbance at 220nm and 275nm.
220nm : to obtain NO₃⁻ reading
275nm : to determine interference due to dissolved organic matter

Calculation:

For samples, blank and std. solutions, subtract two times the absorbance reading at 275nm from the reading at 220nm to obtain absorbance due to NO₃⁻:

$$2DS = Abs_{220} - 2 * Abs_{275}$$

where:

- 2DS = absorbance due to NO₃⁻
- Abs₂₂₀ = the absorbance reading at 220nm
- Abs₂₇₅ = the absorbance reading at 275nm.

Construct a standard curve by plotting absorbance due to NO₃⁻(2DS) against NO₃⁻-N concentration of standard. Using corrected sample absorbances, obtain sample concentrations directly from standard curve.

TN, DTN

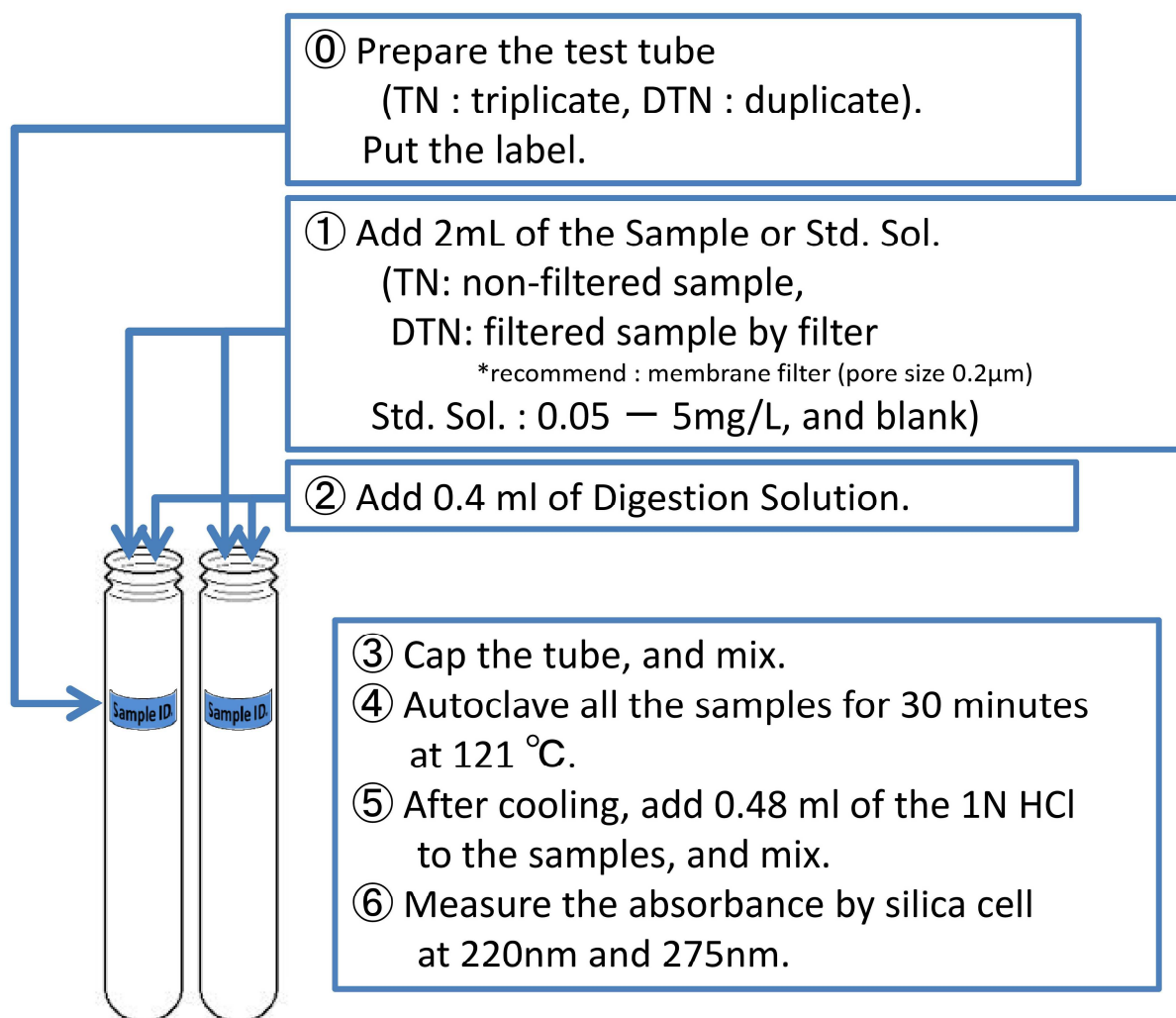


Fig. 1. Flow chart of the analytical procedure for TN/DTN.

③ Nitrate Analysis by Ultraviolet Spectrophotometric Screening Method

(only in the case of low organic matter contents)

(ref. : Standard methods 22nd edition 4500-NO₃-B)

*volume is different

Equipment:

- 200ml measuring cylinder
- transfer pipet
- pipette(10ml, 1ml)
- bottle
- tubes with cap
- silica cuvette(1cm) : *cannot use glass cuvette, that block UV light.
- syringe
- syringe filter (0.2μm) : i.e. cellulose acetate

Apparatus:

- Analytical balance
- Spectrophotometer

Interference:

Dissolved organic matter, surfactants, NO₂⁻, and Cr⁶⁺ interfere. Various inorganic ions not normally found in natural water, such as chlorite and chlorate, may interfere. inorganic substances can be compensated for by independent analysis of their concentrations and preparation of individual correction curves.

Required Chemicals:

1. Freshly prepared nitrate free water
2. 10 ml, Conc. HCl : G.R.
3. Nitrate Nitrogen Standard Solution (1000mg- NO₃-N/l)

Preparation of color reagent:

1. Hydrochloric acid solution, HCl, 1N : (0.1ml/tube)
To 110ml of distilled water, add 10 ml of Hydrochloric Acid
2. Nitrate Standard solutions: (0.05 ~ 5 mg/l)
See Table.1

Procedure: see Fig. 2

a. **Removal of suspended particles:**

Filter through a syringe filter (0.2µm)

b. **Sample Preparation:**

duplicate all the samples

Dilute the sample (if required)

* had better check nitrate concentration by pack test.

To 5 ml of sample, add 0.1 ml of Hydrochloric acid solution, mix thoroughly.

c. **Standard solutions preparation:**

duplicate blank and all the standard solutions

To 5 ml of blank and std. solutions, add 0.1 ml of Hydrochloric acid solution, mix thoroughly.

d. **Photometric measurement:**

Measure the absorbance at 220nm and 275nm.

220nm : to obtain NO₃⁻ reading

275nm : to determine interference due to dissolved organic matter

Calculation:

For samples, blank and std. solutions, subtract two times the absorbance reading at 275nm from the reading at 220nm to obtain absorbance due to NO₃⁻:

$$2DS = Abs_{220} - 2 * Abs_{275}$$

where:

2DS = absorbance due to NO₃⁻

Abs₂₂₀ = the absorbance reading at 220nm

Abs₂₇₅ = the absorbance reading at 275nm.

Construct a standard curve by plotting absorbance due to NO₃⁻(2DS) against NO₃⁻-N concentration of standard. Using corrected sample absorbances, obtain sample concentrations directly from standard curve.

NO₃-N

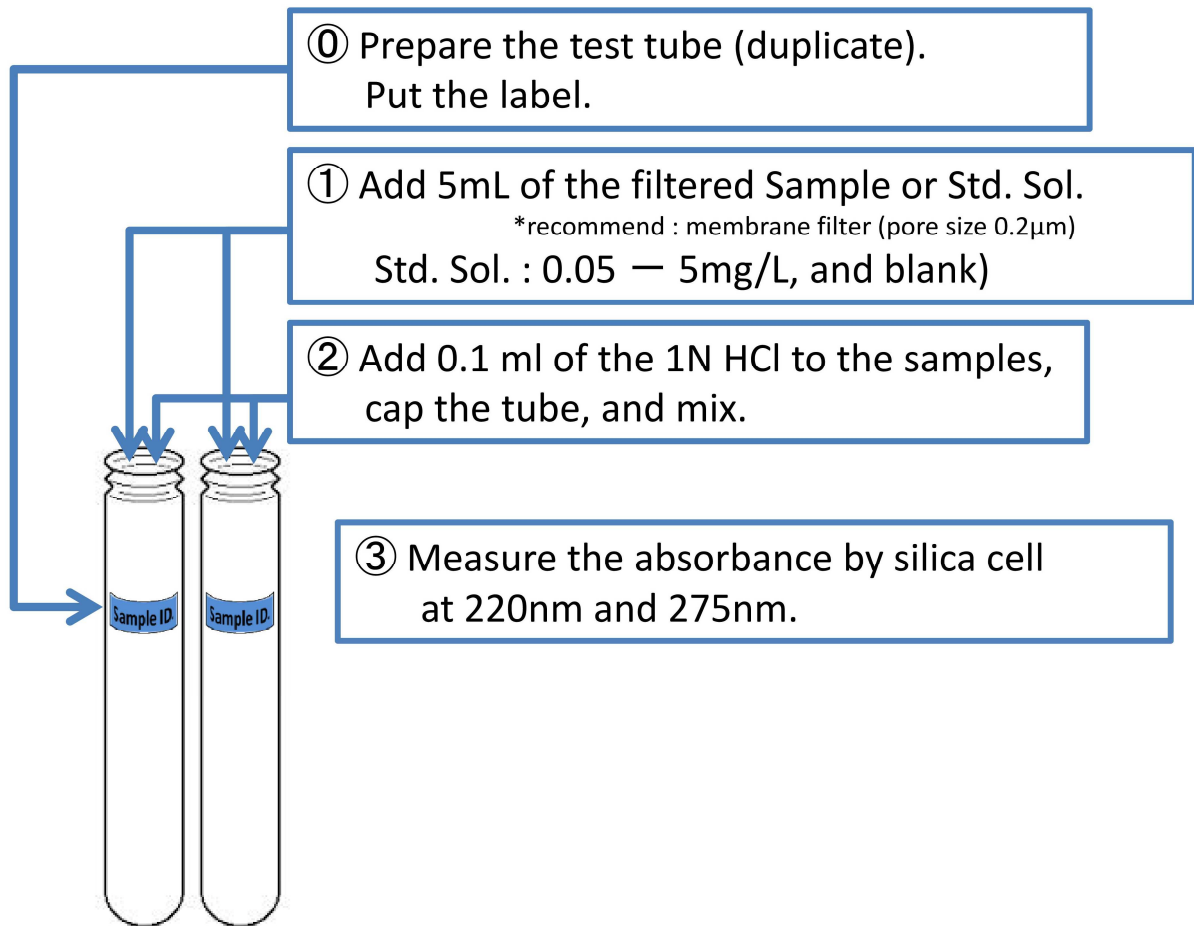


Fig. 2. Flow chart of the analytical procedure for NO₃.

③-2 Nitrate Analysis by Second-Derivative Ultraviolet Spectrophotometric Method

(ref. : Standard methods 22nd edition 4500-NO₃-C)

*volume is different

Equipment:

- transfer pipet
- pipette(10ml, 1ml)
- bottle
- tubes with cap
- silica cuvette(1cm) : *cannot use glass cuvette, that block UV light.
- syringe
- syringe filter (0.2μm) : i.e. cellulose acetate

Apparatus:

- Spectrophotometer

Interference:

The nitrate UV spectrum is similar to that of nitrate. However, nitrite concentrations usually are much lower than nitrate concentrations. Bicarbonate absorbs weakly at wavelengths below 210 nm, but does not affect the second-derivative signal of nitrate. Bromide interferes at seawater concentrations (68 mg Br-/L, salinity 35%) so this method cannot be used to determine nitrate in seawater. Neither Fe nor Cu interferes at 2mg/L but both metals seriously interfere at 20mg/L. The method has been tested only for potable water. Its suitability for nitrate determination in seawater has not been tested.

Required Chemicals:

4. Freshly prepared nitrate free water
5. 10 ml, Conc. HCl : G.R.
6. Nitrate Nitrogen Standard Solution (1000mg- NO₃-N/l)

Preparation of color reagent:

1. Hydrochloric acid solution, HCl, 1N : (0.1ml/tube)
To 110ml of distilled water, add 10 ml of Hydrochloric Acid
2. Nitrate Standard solutions: (5 ~ 25 mg/l)
See Table.2

Procedure: see Fig. 3

- a. **Removal of suspended particles:**
Filter through a syringe filter (0.2µm)
- b. **Check the absorbance of sample:**
Pipet 5.0 ml sample into the tube, add 0.1ml of 1N HCl, and shake
Scan from 250 nm to 200 nm, and check the maximum absorbance point in the range 230 to 220 nm
* if absorbance is beyond 0.5 Abs, dilute sample with nitrate-free water
- c. **Sample preparation:**
Put in 9 ml the sample (or diluted sample) into the tubes, 0.2 ml 1N HCl, 1 ml standard solution to tubes
* standard solution : blank, 1, 2, 3, 4, 5 mg-N/ml
each concentration prepare duplicate tubes
- d. **Photometric measurement:**
Measure the absorbance every 1.0nm in the range 231 to 219 nm, and record each value

Calculation:

- (1) For each tube, compute second-derivative spectrum by above absorbance of each wavelength, and find maximum values (see Fig.4)
- (2) Use the simplified least-squares procedure to simultaneously smooth and differentiate spectra
Perform least-squares linear regression using the second derivatives of the blank and standard spectra
Sample concentration is (Fig.4):

$$C(\text{mg} - \text{N/l}) = \frac{\text{Int}}{\text{Slp}} \times \frac{1}{V}$$

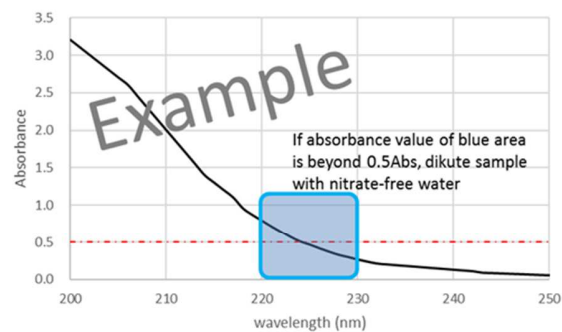
where:

- Slp : slope of regression line,
- Int : intercept of regression line, and
- V : sample volume (ml).

NO₃-N (Second-derivative method)

1. Check the absorbance of sample

Pipet 5mL filtered sample and 0.1mL of 1N HCl
Scan from 250 to 200nm



2. Measurement

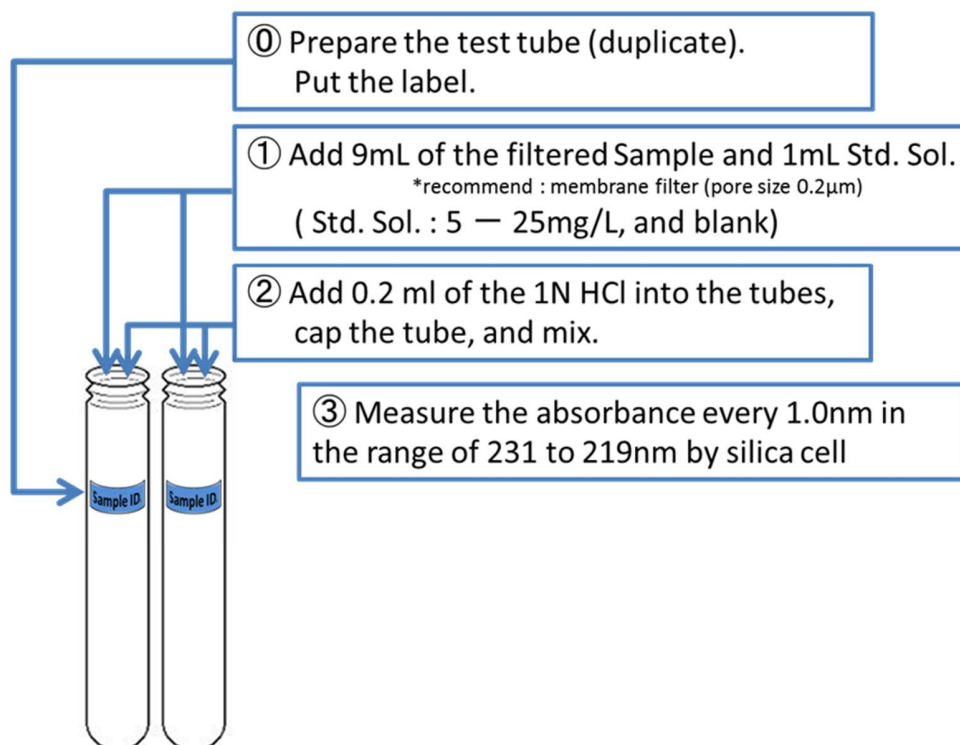


Fig. 3. Flow chart of NO₃ second derivative method

NO₃-N (Second-derivative method)

3. Calculation

(1) Second-derivative

For each tube, compute second-derivative spectrum by above absorbance of each wavelength, and find maximum values

Absorbance	add NO ₃ -N (μg)					
WL(nm)	2.5	2	1.5	1	0.5	0
219	0.8680	-	-	-	-	0.1204
220	0.7902	-	-	-	-	0.1176
221	0.7117	-	-	-	-	0.1155
229	0.2981	-	-	-	-	0.0995
230	0.2650	-	-	-	-	0.0972
231	0.2371	-	-	-	-	0.0956

← actual concentration

— Absorbance values

Ex) second-derivative at X nm

$$2DV_x = \frac{\left(\frac{Abs_{x+1} - Abs_x}{WL_{x+1} - WL_x}\right) - \left(\frac{Abs_x - Abs_{x-1}}{WL_x - WL_{x-1}}\right)}{(WL_{x+1} - WL_{x-1})} = \frac{Abs_{x+1} - 2 * Abs_x + Abs_{x-1}}{(WL_{x+1} - WL_{x-1})}$$

2nd derivative	add NO ₃ -N (μg)					
WL(nm)	2.5	2	1.5	1	0.5	0
220	-0.0004	-	-	-	-	0.0004
230	0.0026	-	-	-	-	0.0004

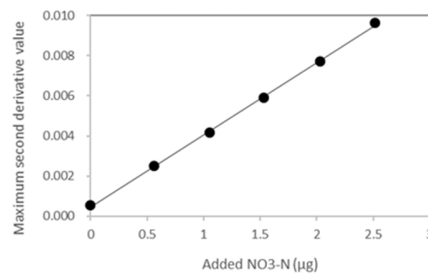
Where:

DV : second-derivative value

Abs_{x+1}, Abs_x, Abs_{x-1} : absorbance at X+1, X, X-1 nm

WL_{x+1}, WL_x, WL_{x-1} : Wavelength

(2) Make a least-squares linear regression



$$S = Int + A \times Slp$$

Where:

S : maximum second-derivative value

A : amount of added NO₃-N (μg)

Int : intercept of regression line

Slp : slope of regression line

(3) Calculate the concentration

$$C = \frac{Int}{Slp} \times \frac{1}{V}$$

Where:

C : concentration (mg/L)

Int, Slp : intercept and slope of above line

V : sample volume (ml)

Fig. 4. Flow chart(2) of NO₃ second derivative method

④ Nitrite Analysis by Colorimetric Method

(ref. : Standard methods 22nd edition 4500-NO₂-B)

***volume is different**

Equipment:

- volumetric flask (100ml, 50ml)
- transfer pipet
- 100ml measuring cylinder
- pipette(10ml, 1ml)
- bottle
- 100 ml Beaker
- tubes with cap
- glass cuvette(1cm)
- syringe
- syringe filter (0.2μm) : i.e. cellulose acetate

Apparatus:

- Analytical balance
- Spectrophotometer

Required chemicals:

1. Freshly prepared nitrite free water
2. 10 ml, Phosphoric Acid (85%) : E.P.
3. 1 gm, Sulfanilamide : G.R.
4. 0.1 gm, N-(1-Naphthyl) ethylenediamine dihydrochloride : G.R.
5. Nitrite Nitrogen Standard Solution (1000mg- NO₂-N/l)

Reagents

1. color reagent : (0.2mL/tube)

To approximately 80ml of distilled water, add 10 ml of Phosphoric Acid (85%), 1 gm Sulfanilamide and 0.1 gm N-(1-Naphthyl) ethylenediamine dihydrochloride and make the total volume of 100 ml.

**** Store the color reagent in dark glass bottle and keep in refrigerator.**

***Note: This color reagent can be used for a month.**

2. Nitrite Standard solutions: (0.02~0.5 mg/l)

See Table.3

Procedure: see Fig. 5

a. **Removal of suspended particles:**

Filter through a syringe filter (0.2 μ m)

b. **Sample Preparation:**

duplicate all the samples

Dilute the sample (if required)

* had better check nitrite concentration by pack test.

To 5 ml of sample, add 0.2 ml of color reagent, mix thoroughly, and leave for 20 minutes in room temperature.

c. **blank and Standard solutions preparation:**

duplicate blank and all standard solutions

To 5 ml of blank and std. solutions, add 0.2 ml of color reagent, mix thoroughly, and leave for 20 minutes in room temperature.

d. **Photometric measurement:**

Measure the absorbance at 543nm.

Calculation:

Prepare a std. curve by plotting absorbance of std. against NO₂⁻-N concentration.
Compute sample concentration from the curve.

**The std. solution has certain absorbance and develops the color after adding color reagent. But sometimes, the samples have higher absorbance than the std. solution but results no color development even after adding color reagent. In such a situation, we have to measure the absorbance of the sample without color reagent and reduce the absorbance value from the sample with color reagent. We use this deducted absorbance as a reading.

Note: The disappearance of the color might be from the organic material present in the sample.

NO₂-N

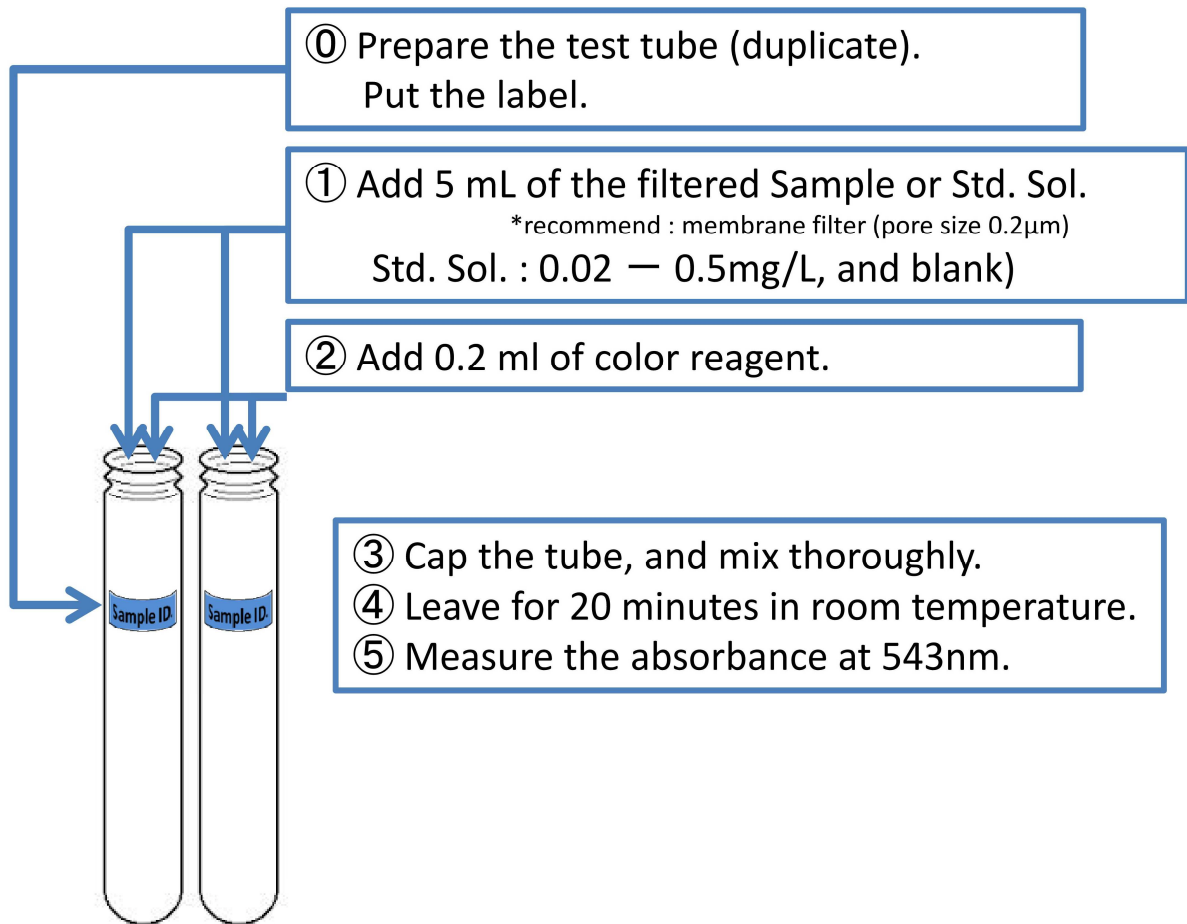


Fig. 5. Flow chart of the analytical procedure for NO₂.

⑤ Ammonia Analysis by Phenate Method

(ref. : Standard Methods for the Examination of Tap Water in Japan)

***volume is different**

Equipment:

- volumetric flask (500ml, 100ml, 50ml)
- transfer pipet
- pipette(10ml, 1ml)
- bottle
- 100 ml Beaker
- tubes with cap
- glass cuvette(1cm)
- syringe
- syringe filter (0.2 μ m) : i.e. cellulose acetate

Apparatus:

- Analytical balance
- Spectrophotometer

Interference:

If hydrogen sulfide is present, remove by acidifying samples to pH 3 with dilute HCl and aerating vigorously until sulfide odor no longer can be detected.

Required Chemicals:

1. Distilled water
2. 5 gm Phenol : G.R.
3. 0.025 gm, Sodium Nitroprusside : G.R.
4. 10 ml, Sodium Hypochlorite (5%) : Chemically Pure
***If fresh Sodium Hypochlorite then, it is 12%**
5. 15 gm, Sodium Hydroxide : for analysis of Nitrogen
6. Ammonium Nitrogen Standard Solution (1000mg-NH₄-N/l)

Reagent:

1. Phenol/Sodium nitroprusside solution (Solution A): (1ml/tube)

To approximately 400 ml water, add 5 gm of Phenol, 0.025 gm of Sodium Nitroprusside and dilute to 500 ml.

****Prepare this solution one day before use/ analysis, to make phenol dissolve**

****Store this solution in dark bottle. This Solution can be used for a month.**

2. Sodium hypochlorite solution (effective chlorine concentration: 0.1w/v%) (Solution B):
(1ml/tube)

Dissolve commercial sodium hypochlorite solution (50/C mL, C :effective chlorine concentration) and 7.5 gm of Sodium Hydroxide in 100ml water and dilute to 500 ml.

**** Store this solution in dark bottle, and refrigerate.**

*** This solution can be used in a range of the effective chlorine concentration (0.05-0.1w/v%).**

3. Ammonium Nitrogen Standard Solution: (0.05 ~ 5 mg/l)

See Table.4

Procedure: see Fig. 6

a. Removal of suspended particles:

Filter through a syringe filter (0.2 μ m)

b. Sample Preparation:

duplicate all the samples

i. Take 2 ml of samples.

(Dilute the sample (if required), and make the final volume of 2 ml in a tube.)

* had better check ammonium concentration by pack test.

ii. Add 1 ml of solution A, to all samples blank and std. solution; and shake gently so that no bubbles are formed.

iii. Add 1 ml of solution B and gently shake, after cap.

iv. Leave the samples for 1 hour at room temperature (20-40°C)

v. After an hour, bluish green color develops.

c. Standard solutions preparation:

duplicate blank and all the standard solutions

Treat blank and std. solutions in same manner as samples.

d. Photometric measurement:

Measure the absorbance at 640nm.

Calculation:

Prepare a std. curve by plotting absorbance of std. against NH₃-N concentration.
Compute sample concentration from the curve.

NH₄-N

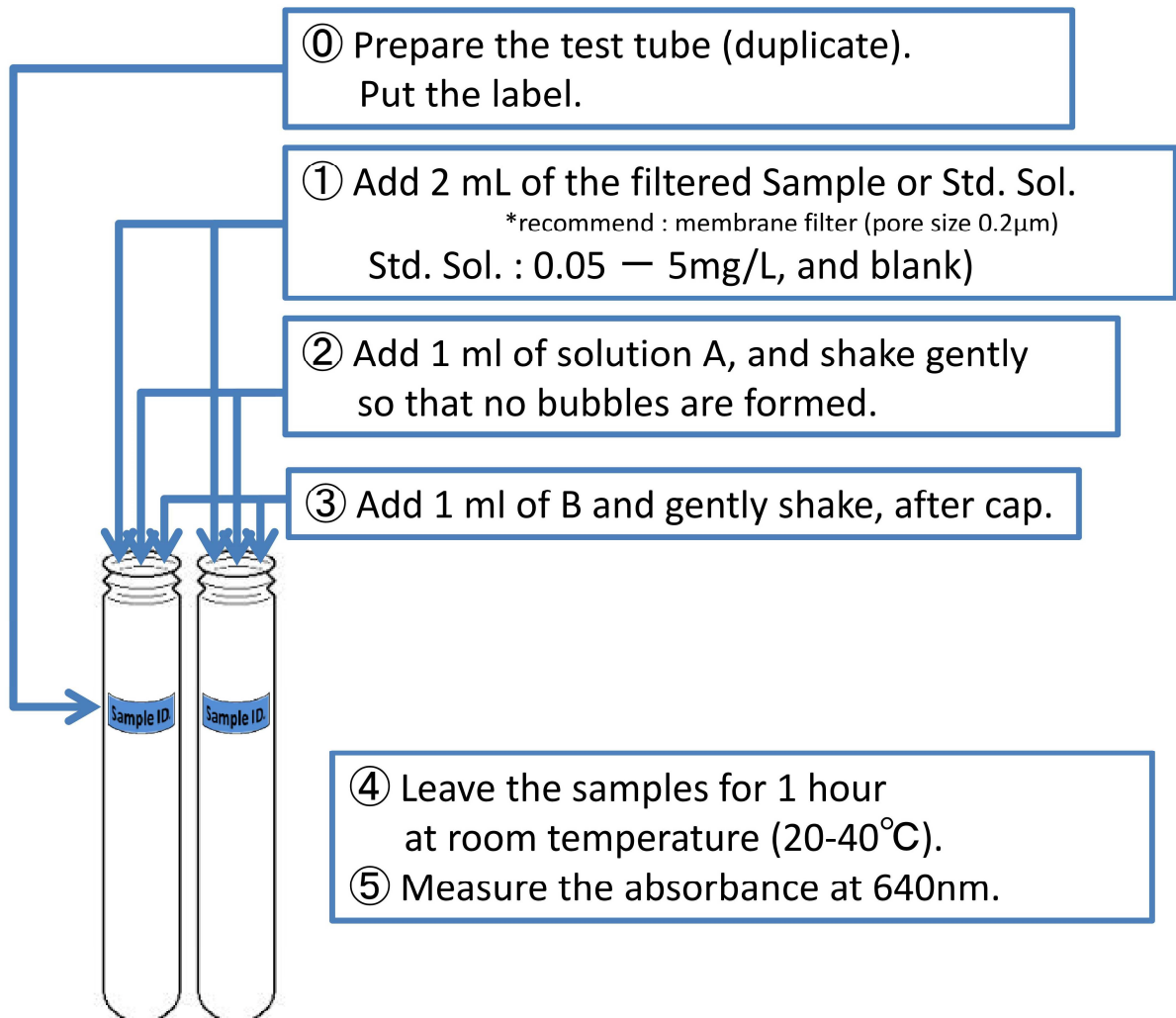


Fig. 6 Flow chart of the analytical procedure for NH₄.

⑥ Total Phosphorus Analysis by Persulfate Digestion Method (ref. : Standard Methods for the Examination of Tap Water in Japan)

***volume is different**

Equipment:

- volumetric flask(500ml, 100ml)
- pipette(10ml, 5ml)
- measuring cylinder(200ml, 50ml)
- bottle
- 100 ml Beaker
- tubes with cap
- glass cuvette(1cm)

Apparatus:

- Magnetic stirrer
- Analytical balance
- Spectrophotometer

Required Chemicals:

1. Distilled water
2. 4 gm, Potassium persulfate (freshly prepared) : for analysis of Phosphorus
3. 50 ml, Conc. H_2SO_4 : G.R.
4. 0.24 gm Potassium antimonyl tartrate: G.R.
5. 6gm, Ammonium molybdate : G.R.
6. gm Ascorbic Acid : G.R.
7. Phosphorus Standard Solution (1000mg- PO_4 -P/l)

Preparation of reagents:

1. 4% Potassium persulfate Solution (Digestion Solution): (0.4ml/tube)
***Total Phosphorus are converted to orthophosphate.**
Prepare Potassium persulfate 4% Weight by Volume. Add 4 gm of Potassium persulfate to 100 ml distilled water. Then stir it with the help of magnetic stirrer.
2. Sulfuric Acid (Solution A): same of orthophosphate
To approximately 100 ml water, add **50 ml** of Conc. H_2SO_4 .
3. Potassium antimonyl tartrate solution (Solution B):
To approximately 300 ml water, add 0.24 gm of Potassium antimonyl tartrate, 6 gm of Ammonium molybdate, and dissolve. Then, add 120mL of Solution A. Let it cool and then make a final volume of 500 ml in a volumetric flask.
4. L-ascorbic Acid (Solution C): same of orthophosphate
To **100 ml** of distilled water, add **7.2 gm** of L-ascorbic Acid
****This reagent can be used only for a week, even refrigerated.**
****It is colorless when freshly prepared and color changes to light yellow when kept for long time)**
5. Combined reagent (Solution D): same of orthophosphate (0.2ml/tube)
Combine Solution B and C in the ration 5:1

6. Phosphorus Standard Solutions: (0.01 ~ 5 mg/l) same of orthophosphate
See Table.5

Procedure: see Fig. 7

a. Sample Preparation:

- i. triplicate all the samples
- ii. Take 2ml of samples.
(Dilute the sample (if required) and take 2 ml of each)
* had better check Phosphorus concentration by pack test.
- iii. Add 0.4 ml of Digestion Solution to each sample.
- iv. Autoclave all the samples for 30 minutes at 121°C.
- v. After cooling, add 0.2ml of the Solution D to the samples.
- vi. Leave the samples for 15 minutes at room temperature (20-40°C), before taking the readings.

b. Standard solutions preparation:

- duplicate blank and all the standard solutions
Treat blank and std. solutions in same manner as samples.

c. Photometric measurement:

- Measure the absorbance at 880nm.

Calculation:

Prepare a std. curve by plotting absorbance of std. against P concentration. Compute sample concentration from the curve.

⑦ Dissolved Phosphorus Analysis by Persulfate Digestion Method

(ref. : Standard Methods for the Examination of Tap Water in Japan)

*volume is different

Equipment:

- volumetric flask(500ml, 100ml)
- pipette(10ml, 5ml)
- measuring cylinder(200ml, 50ml)
- bottle
- 100 ml Beaker
- tubes with cap
- glass cuvette(1cm)
- syringe
- syringe filter (0.2 μ m) : i.e. cellulose acetate

Apparatus:

- Magnetic stirrer
- Analytical balance
- Spectrophotometer

Required Chemicals:

1. Distilled water
2. 4 gm, Potassium persulfate (freshly prepared) : for analysis of Phosphorus
3. 50 ml, Conc. H₂SO₄ : G.R.
4. 0.24 gm Potassium antimonyl tartrate: G.R.
5. 6gm, Ammonium molybdate : G.R.
6. 7.2 gm Ascorbic Acid : G.R.
7. Phosphorus Standard Solution (1000mg-PO₄-P/l)

Preparation of reagents:

1. 4% Potassium persulfate Solution (Digestion Solution): same of Total Phosphorus (0.4ml/tube)

*Dissolved Phosphorus are converted to orthophosphate.

Prepare Potassium persulfate 4% Weight by Volume. Add 4 gm of Potassium persulfate to 100 ml distilled water. Then stir it with the help of magnetic stirrer.

2. Sulfuric Acid (Solution A): same of orthophosphate

To approximately 100 ml water, add 50 ml of Conc. H₂SO₄.

3. Potassium antimonyl tartrate solution (Solution B):

To approximately 300 ml water, add 0.24 gm of Potassium antimonyl tartrate, 6 gm of Ammonium molybdate, and dissolve. Then, add 120mL of Solution A. Let it cool and then make a final volume of 500 ml in a volumetric flask.

4. L-ascorbic Acid (Solution C): same of orthophosphate
To 100 ml of distilled water, add 7.2 gm of L-ascorbic Acid
****This reagent can be used only for a week, even refrigerated.**
****It is colorless when freshly prepared and color changes to light yellow when kept for long time)**
5. Combined reagent (Solution D): same of orthophosphate (0.2ml/tube)
Combine Solution B and C in the ration 5:1
6. Phosphorus Standard Solutions: (0.01~1.0 mg/l) same of orthophosphate
See Table.5

Procedure: see Fig. 7

- a. Removal of suspended particles:
Filter through a syringe filter (0.2 μ m)
- b. Sample Preparation:
 - i. duplicate all the samples
 - ii. Take 2ml of samples. (Dilute the sample (if required) and take 2 ml of each)
* had better check Phosphorus concentration by pack test.
 - iii. Add 0.4 ml of Digestion Solution to each sample.
 - iv. Autoclave all the samples for 30 minutes at 121°C.
 - v. After cooling, add 0.2ml of the Solution D to the samples.
 - vi. Leave the samples for 15 minutes in room temperature(20-40°C), before taking the readings.
- b. Standard solutions preparation:
duplicate blank and all the standard solutions
Treat blank and std. solutions in same manner as samples.
- c. Photometric measurement:
Measure the absorbance at 880nm.

Calculation:

Prepare a std. curve by plotting absorbance of std. against P concentration. Compute sample concentration from the curve.

TP, DTP

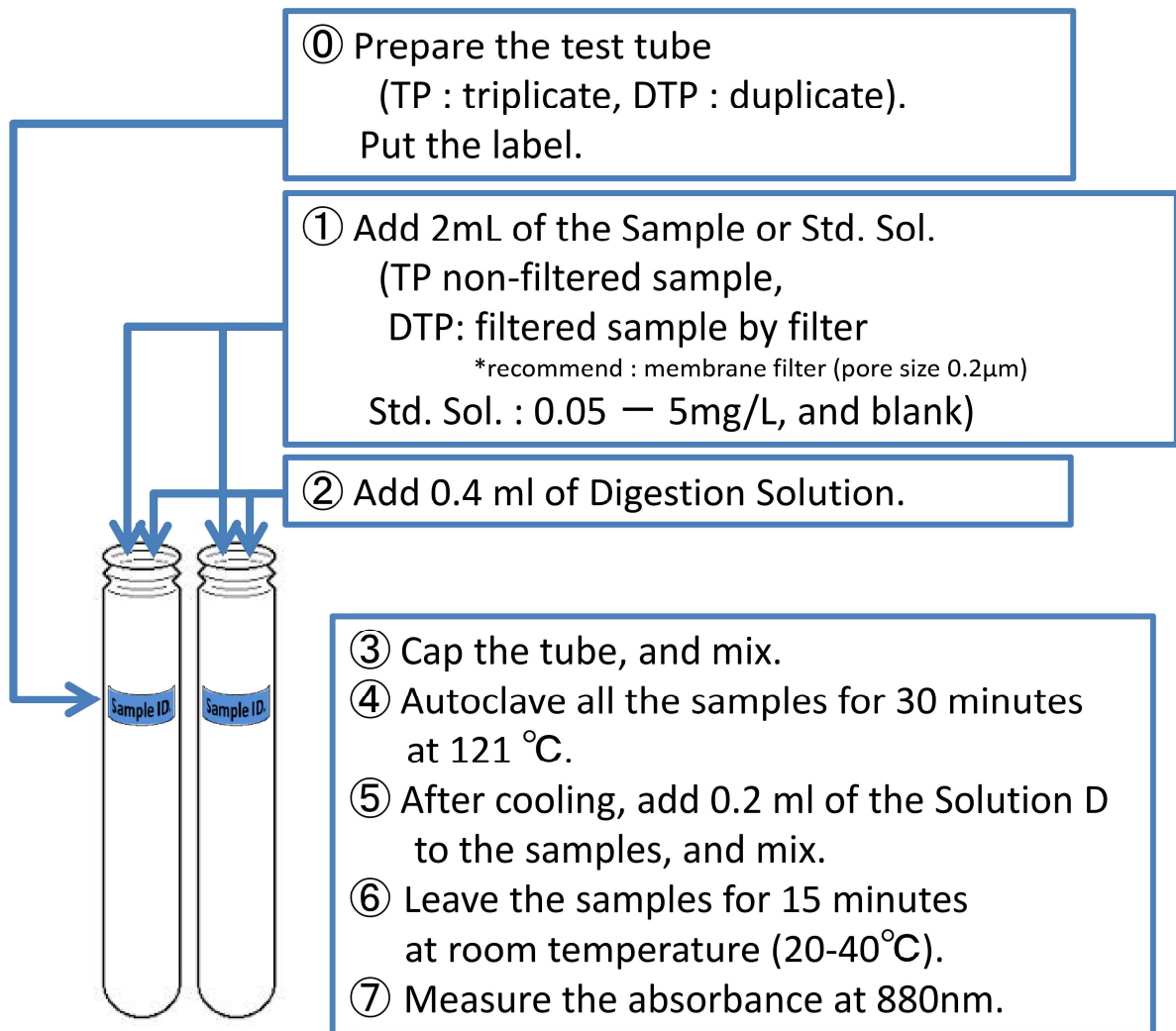


Fig. 7 Flow chart of the analytical procedure for TP/DTP.

⑧ Orthophosphate Analysis by Ascorbic Acid Method

(ref. : Standard Methods for the Examination of Tap Water in Japan)

***volume is different**

Equipment:

- volumetric flask(500ml, 100ml)
- transfer pipet
- pipette(10ml, 5ml)
- measuring cylinder(200ml, 50ml)
- bottle
- 100 ml Beaker
- tubes with cap
- glass cuvette(1cm)
- syringe
- syringe filter (0.2 μ m) : i.e. cellulose acetate

Apparatus:

- Magnetic stirrer
- Analytical balance
- Spectrophotometer

Interference:

ferric ion (1mg/L<)

Required Chemicals:

1. Distilled water
2. 50 ml, Conc. H₂SO₄ : G.R.
3. 0.24 gm Potassium antimonyl tartrate: G.R.
4. 6gm, Ammonium molybdate : G.R.
5. 7.2 gm Ascorbic Acid : G.R.
6. Phosphorus Standard Solution (1000mg-PO₄-P/l)

Preparation of reagents:

1. Sulfuric Acid (Solution A):

To approximately 100 ml water, add 50 ml of Conc. H₂SO₄.

2. Potassium antimonyl tartrate solution (Solution B):

To approximately 300 ml water, add 0.24 gm of Potassium antimonyl tartrate, 6 gm of Ammonium molybdate, and dissolve. Then, add 120mL of Solution A. Let it cool and then make a final volume of 500 ml in a volumetric flask.

3. L-ascorbic Acid (Solution C):

To 100 ml of distilled water, add 7.2 gm of L-ascorbic Acid.

****This reagent can be used only for a week, even refrigerated.**

****It is colorless when freshly prepared and color changes to light yellow when kept for long time)**

4. Combined reagent (Solution D): (0.2ml/tube)
Combine Solution B and C in the ration 5:1
5. Orthophosphate Standard Solutions: (0.01~1.0 mg/l)
See Table.5

Procedure: see Fig. 8

- a. Removal of suspended particles:
Filter through a syringe filter (0.2 μ m)
- b. Sample Preparation:
duplicate all the samples
Dilute the sample (if required)
* had better check Phosphorus concentration by pack test.
To 2 ml of blank and std. solutions, add 0.2 ml of Solution D, mix thoroughly, and leave for 15 minutes in room temperature(20-40°C).
- c. Standard solutions preparation:
duplicate blank and all the standard solutions
Treat blank and std. solutions in same manner as samples.
- d. Photometric measurement:
Measure the absorbance at 880nm.

Calculation:

Prepare a std. curve by plotting absorbance of std. against PO₄ concentration. Compute sample concentration from the curve.

PO₄

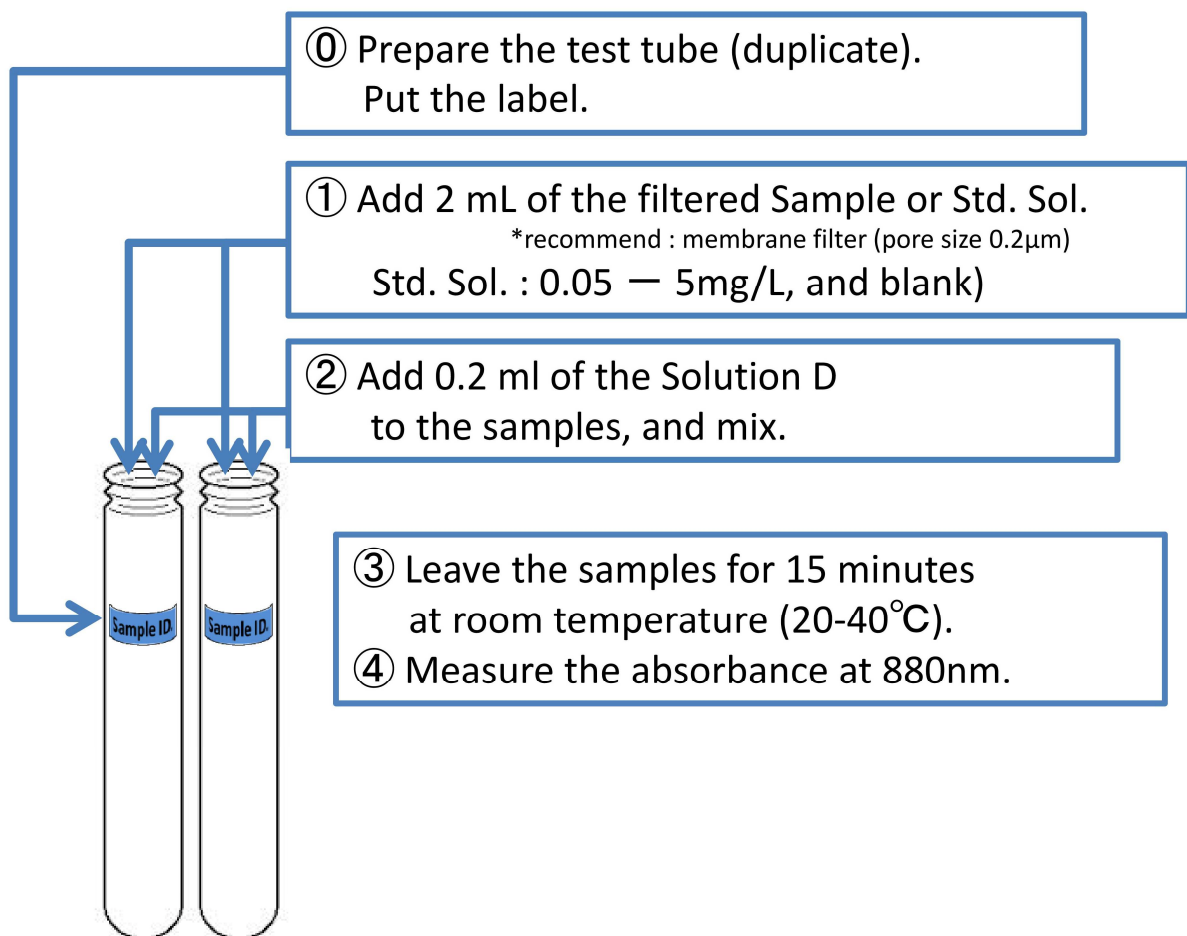


Fig. 8 Flow chart of the analytical procedure for PO₄.

⑨ Total iron by Phenanthroline Method

(ref. : Standard Methods 22nd edition 3500-Fe B)

Required Apparatus:

1. Volumetric flask (20ml, 100ml)
2. 100ml measuring cylinder
3. Pipette (10ml and 1ml)
4. Screw bottle (30 mL)
5. 100ml beaker
6. Analytical balance
7. Spectrophotometer
8. Glass cuvette (1.5ml)
9. Syringe
10. Syringe filter (0.2 μm): cellulose acetate
11. Hot plate
12. Teflon beaker

Required chemicals:

1. Freshly prepared nitrite free water
2. Conc. Hydrochloric acid
3. Hydroxylamine Hydrochloride ($\text{NH}_2\text{OH}\cdot\text{HCL}$)
4. Ammonium Acetate ($\text{NH}_4\text{C}_2\text{H}_3\text{O}_2$)
5. 1,10-phenanthroline monohydrate ($\text{C}_{12}\text{H}_8\text{N}_2\cdot\text{H}_2\text{O}$)
6. Iron Stock Solution

Preparation of color reagent:

Store reagents in glass-stoppered bottles.

The hydroxylamine, phenanthroline, and stock iron solutions are stable for several months. The standard iron solutions are not stable; prepare daily as needed by diluting the stock solution.

Visual standards in nessler tubes are stable for several months if sealed and protected from light

1. Hydrochloric acid, HCl, conc, containing less than 0.5 ppm iron:

Add 5ml of Hydrochloric acid, to 55 ml of distilled water.

*This solution is stable indefinitely if tightly stoppered.

2. Hydroxylamine solution:

Dissolve 10g Hydroxylamine Hydrochloride in 100 mL water.

*This solution is stable for several months

3. Ammonium acetate buffer solution:

Dissolve 250 g Ammonium Acetate in 150 mL water. Add 700 mL conc (glacial) acetic acid. Because even a good grade of Ammonium Acetate contains a significant amount of iron, prepare new reference standards with each buffer preparation.

* This solution is stable indefinitely if tightly stoppered.

4. Phenanthroline solution:

Dissolve 100 mg 1,10-phenanthroline monohydrate, in 100 mL water by stirring and heating to 80°C. Do not boil. Discard the solution if it darkens. Heating is unnecessary if 2 drops conc HCl are added to the water.

* Note: One milliliter of this reagent is sufficient for no more than 100 µg Fe

5. Iron standard solution (0~1.0mg/L)

See Table.6

Procedure: see Fig. 9

a. Removal of suspended particles:

Filter the sample through a syringe filter (0.2µm)

b. Sample Preparation:

Duplicate all the samples

Mix sample thoroughly and measure 50.0 mL into a 125-mL erlenmeyer flask.

Dilute the sample (if this sample volume contains more than 200µg iron use a smaller accurately measured portion and dilute to 50ml.)

Add 2 mL conc HCl and 1 mL NH₂OH · HCl solution.

Add a few glass beads and heat to boiling.

To insure dissolution of all the iron, continue boiling until volume is reduced to 15 or 20 mL.(if the sample is ashed, take up residue in 2 mL conc HCl and 5 mL water)

Cool room temperature and transfer to 50- or 100-mL volumetric flask.

Add 10 mL NH₄C₂H₃O₂ buffer solution and 4 mL phenanthroline solution, and dilute to mark with water.

Mix thoroughly and allow a minimum of 10 min for maximum color development.

c. Blank and Standard Solutions Preparation:

Duplicate the blank and all standard solutions.

Treating in the same way as samples.

d. Photometric measurement:

Measure the absorbance at 510nm.

Calculation:

Prepare a standard curve by plotting absorbance of standard against Fe concentration. Compute sample concentration from the curve.

$$\text{mg Fe/L} = \frac{\mu\text{g Fe (in 100 mL final volume)}}{\text{mL sample}}$$

* Report details of sample collection, storage, and pretreatment if they are pertinent to interpretation of results.

** The standard solution has certain absorbance and develops the color after adding color reagent. But sometimes, the samples have higher absorbance than the standard solution but results no color development even after adding color reagent. In such a situation, we have to measure the absorbance of the sample without color reagent and reduce the absorbance value from the sample with color reagent. We use this deducted absorbance as a reading.

Note: The disappearance of the color might be from the organic material present in the sample.

T-Fe

Preparation of sample

- ① Prepare the Teflon beaker(duplicate). Put the label.
- ① Measure 50mL sample or Std. Sol. into a Teflon beaker.
(Std. Sol. : 0.2 — 1mg/L, and blank)
- ② Add 2mL conc HCl and 1mL Hydroxylamine solution.
- ③ Add a few glass beads and heat to boil at Hot plate.
- ④ Continue boiling until volume is reduced 15 to 20mL.
- ⑤ Cool to room temperature.

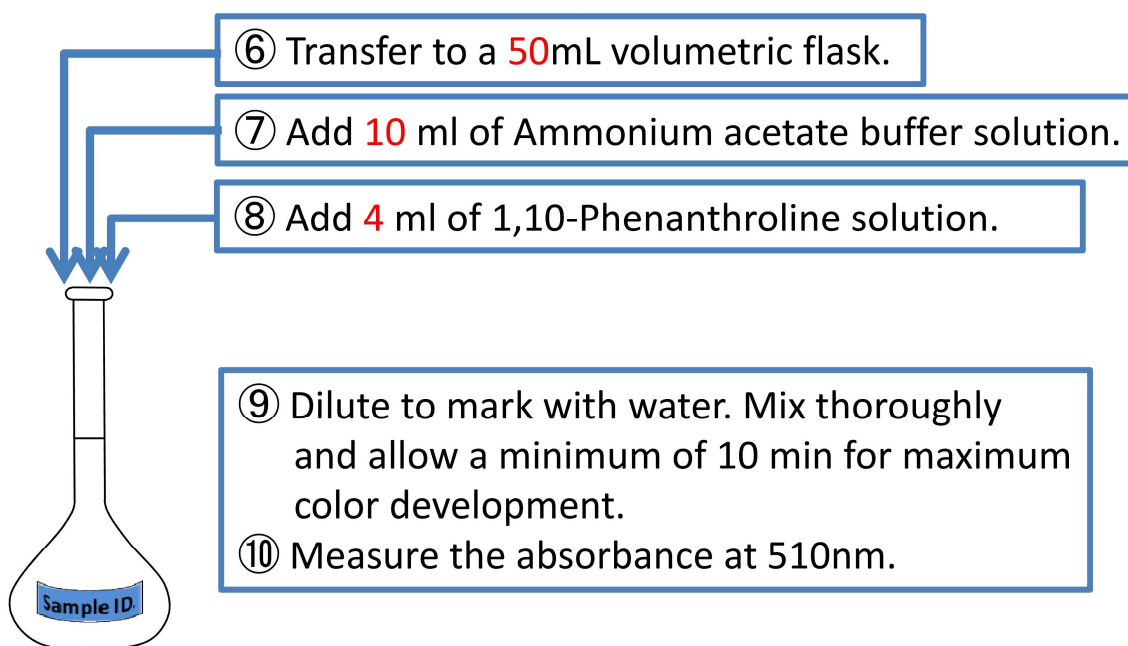


Fig. 9 Flow chart of the analytical procedure for T-Fe.

⑩ Acid soluble iron by Phenanthroline Method (ref. : water quality research method (Japan))

***volume is different**

Equipment:

- measuring cylinder (200mL, 1L)
- volumetric flask (20ml, 1L)
- transfer pipet
- pipette(10ml, 1ml)
- bottle
- 300 ml Beaker
- tubes with cap
- Teflon beaker
- glass cuvette(1cm)

Apparatus:

- Magnetic stirrer
- Analytical balance
- Spectrophotometer

Required Chemicals:

1. Distilled water
2. Conc. HCl : containing less than 0.5 ppm iron
3. 1,10-Phenanthroline monohydrate : G.R.
4. Ammonium acetate : G.R.
5. conc (glacial) Acetic acid : G.R.
6. Hydroxylamine Hydrochloride
7. Ammonium hydroxide(28%)
8. Fe Standard Solution (1000mg- Fe/l)

Preparation of reagents:

1. 1,10-Phenanthroline solution : (1ml/tube)
Dissolve 0.1 gm 1,10-Phenanthroline monohydrate, in 100 ml distilled water by stirring and heating to 80°C (**do not boil**). Heating is unnecessary if 2 drops conc HCl are added to the water.
**** Store this solution in dark bottle, and refrigerate.**
**** Discard the solution if it darkness.**
2. Ammonium acetate buffer solution : (1ml/tube)
Dissolve 250gm Ammonium acetate in 150mL water. Add 700mL conc (glacial) Acetic acid.
3. Preparation of Iron Standard solutions: (0.05~1 mg/l)
See Table.6
4. Hydroxylamine solution : (0.4ml/tube of Std.Sol.)
Dissolve 10g Hydroxylamine Hydrochloride in 100mL water.

5. 3N HCl (1.2ml/each Teflon beaker)
Add 10ml HCl in 30 ml distilled water.

Procedure: see Fig. 10

a. Sample Preparation:

***Sample with 1ml-HCl/100ml-sample at time of collection.**

Fill bottle directly from sampling source and stopper.

- i. Duplicate all the samples
- ii. Take 10ml portion of acidified samples (50 ml Teflon beaker).
(Dilute the sample (if required) and take 10 ml of each)
- iii. Add 1.2 ml of 3N HCl to each sample
- iv. Heat for 5 minutes by hotplate
- v. Then, cool. Transfer to a 20ml volumetric flask (if sample is turbid, need to filter)
- vi. Add 0.4 ml Hydroxylamine solution to each tubes
- vii. Add 1.0 ml 1,10-Phenanthroline solution to each tubes
- viii. Add 4 ml of Ammonium acetate buffer solution with vigorous stirring
- ix. Dilute to 20mL, and mix thoroughly

***Do not expose to sunlight.**

b. Standard solutions preparation:

Duplicate blank and all the standard solutions

Take 10 ml of blank and std. sol.

Add 0.2 ml conc HCL.

Then, treat in same manner as samples (vi-ix).

c. Photometric measurement:

Measure color intensity within 5 to 10 min at 510nm.

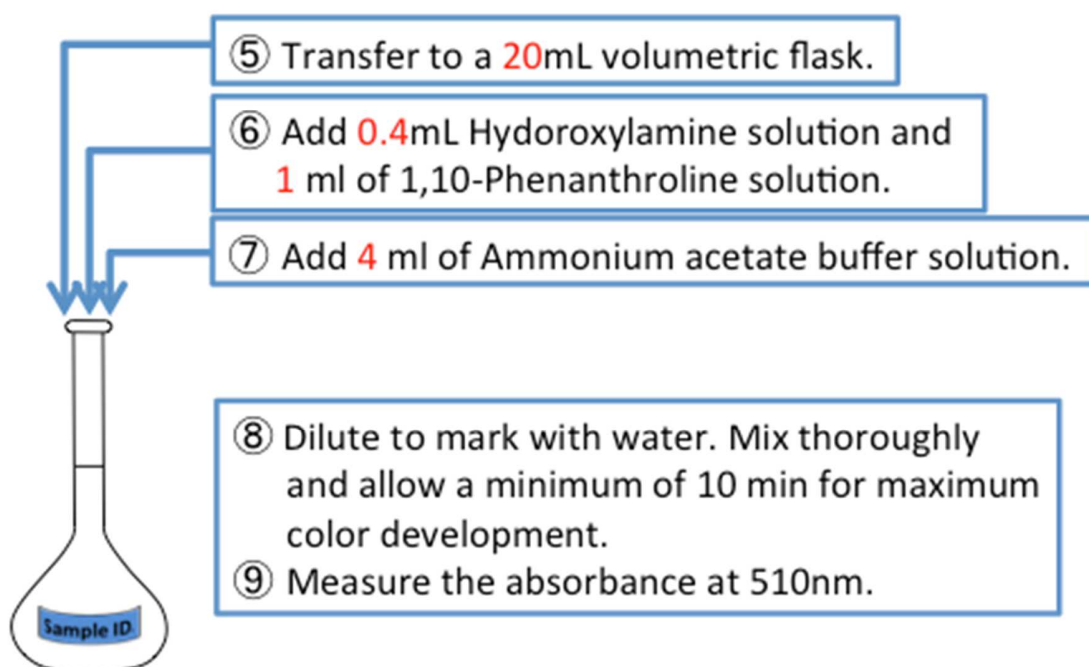
Calculation

Prepare a std. curve by plotting absorbance of std. against ferrous ion concentration.
Compute sample concentration from the curve.

AS-Fe

Preparation of sample

- ① Prepare the Teflon beaker(duplicate). Put the label.
- ① Measure 10mL sample into a Teflon beaker.
(Std. Sol. : 0.2 – 1mg/L, and blank)
- ② Add 1.2mL of 3N HCl.
- ③ Heat for 5minutes by Hot plate.
- ④ Cool to room temperature.



Procedure for Std. Sol.

- (Std. Sol. : 0.2 – 1mg/L, and blank)
- Prepare the test tube(duplicate). Put the label.
 - Add 10mL of the Std. Sol.
 - ⑦ - ⑨

Fig. 10 Flow chart of the analytical procedure for AS-Fe.

⑪ Ferrous ion by Phenanthroline Method

(ref. : Standard Methods 22nd edition 3500-Fe B)

***volume is different**

Equipment:

- measuring cylinder (200mL, 1L)
- volumetric flask (1L)
- transfer pipet
- pipette(10ml, 1ml)
- bottle
- 300 ml Beaker
- tubes with cap
- glass cuvette(1cm)

Apparatus:

- Magnetic stirrer
- Analytical balance
- Spectrophotometer

Required Chemicals:

1. Distilled water
2. Conc. HCl : containing less than 0.5 ppm iron
3. 1,10-Phenanthroline monohydrate : G.R.
4. Ammonium acetate : G.R.
5. conc (glacial) Acetic acid : G.R.
6. Fe Standard Solution (1000mg- Fe/l)

Preparation of reagents:

1. 1,10-Phenanthroline solution : (2ml/tube)
Dissolve 0.1 gm 1,10-Phenanthroline monohydrate, in 100 ml distilled water by stirring and heating to 80°C(**do not boil**). Heating is unnecessary if 2 drops conc HCl are added to the water.
**** Store this solution in dark bottle, and refrigerate.**
**** Discard the solution if it darkness.**
2. Ammonium acetate buffer solution : (1ml/tube)
Dissolve 250gm Ammonium acetate in 150mL water. Add 700mL conc (glacial) Acetic acid.
3. Preparation of Iron Standard solutions: (0.05~1 mg/l)
See Table.6
4. Hydroxylamine solution : (0.1ml/tube of Std.Sol.)
Dissolve 10g Hydroxylamine Hydrochloride in 100mL water.

Procedure: see Fig. 11

b. **Sample Preparation:**

***Sample with 2ml-HCl/100ml-sample at time of collection.**

Fill bottle directly from sampling source and stopper.

- i. Duplicate all the samples
- ii. Take 5ml portion of acidified samples.
(Dilute the sample (if required) and take 5 ml of each)
- iii. Add 2 ml of 1,10-Phenanthroline solution to each sample.
- iv. Add 1 ml of Ammonium acetate buffer solution with vigorous stirring.
- v. Dilute to 10mL, and mix thoroughly.

***Do not expose to sunlight.**

b. **Standard solutions preparation:**

Duplicate blank and all the standard solutions

Add 0.05 ml conc HCL and 0.1ml Hydroxylamine solution to each tubes.

Then, treat in same manner as samples(iii-v).

c. **Photometric measurement:**

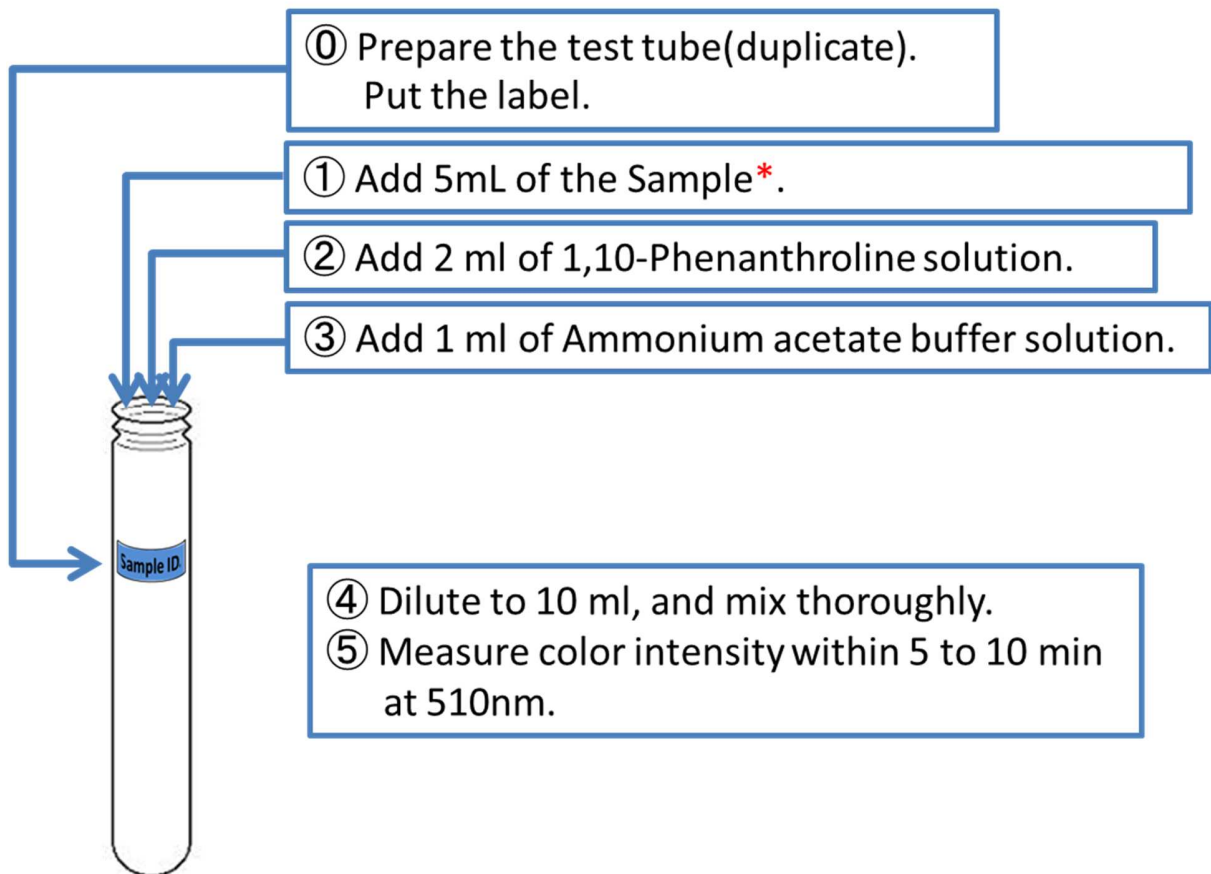
Measure color intensity within 5 to 10 min at 510nm.

Calculation

Prepare a std. curve by plotting absorbance of std. against ferrous ion concentration.
Compute sample concentration from the curve.

Fe²⁺

***Sample with 2ml-HCl/100ml-sample at time of collection.
Fill bottle directly from sampling source and stopper.**



Procedure for Std. Sol.

(Std. Sol. : 0.2 – 1mg/L, and blank)

- ① Prepare the test tube(duplicate). Put the label.
- ② Add 5mL of the Std. Sol.
- ③ Add 0.05mL conc HCl and 0.1mL Hydroxylamine solution.
- ④ Add 2 ml of 1,10-Phenanthroline solution.
- ⑤ Add 1 ml of Ammonium acetate buffer solution.
- ⑥ Dilute to 10 ml, and mix thoroughly.
- ⑦ Measure color intensity within 5 to 10 min at 510nm.

Fig. 11 Flow chart of the analytical procedure for Fe²⁺.

⑫ Dissolved Manganese by Absorptiometric method (Holm aldoxime method) (ref. : Analytical method of mineral spring in Japan)

Equipment:

- Pipette (1,210 mL)
- Screw bottle (30 mL)
- Beaker
- Tubes and cap
- Volumetric flask(100,200mL,1L)
- Screw bottle (30ml)
- Analytical balance
- UV Spectrophotometer
- glass cuvette (1.5ml)

Required Chemicals:

1. Holm aldoxime reagent
2. buffer solution (pH = 10)
3. L- ascorbic acid sodium salt
4. 0.1M EDTA solution
5. Manganese stock solution (1000mg-Mn²⁺/L)

Preparation of reagents:

1. Holm aldoxime reagent
Approximately 100 mL of distilled water , add 8 g of Hydroxylamine hydrochloride (NH₂-OH · HCl). After that add 4ml of 37% formaldehyde (HCHO), adjust the volume of 200ml with distilled water.
2. buffer solution (pH = 10)
Approximately 300 mL of distilled water , add 68 g of Ammonium chloride (NH₄Cl). After that add 570 mL of conc. NH₃ solution, adjust the volume of 1L with distilled water.
3. 0.1M EDTA solution
Dissolve 3.7g of Disodium EDTA dihydrate [(CH₂COO)₂N · CH₂ · CH₂N (CH₂COO)₂H₂ · Na₂ · 2H₂O] in distilled water adjust the final volume of 100mL.
4. Manganese standard solution (0~5.0mg/L)
See Table.7

Procedure: see Fig. 12

a.) Suspended particles Removal

- Filter through a syringe filter (0.2 μm)

b.) Sample preparation

- Duplicate all the samples
- Take 20 mL of samples. (Dilute the sample {if required})
- Add 1 mL of Holm aldoxime reagent and 0.1M EDTA solution of each tube and close with cap.
- Mix and Leave the samples for 5 minutes by control condition at room temperature.
- As the 20 ~ 27°C, stir in addition L- ascorbic sodium salt 10mg and a 0.1M EDTA 1mL.

*The order of addition of sodium L- ascorbic acid → EDTA.
- Leave the samples for 10 minutes by control condition at room temperature.
- Measure absorbance by ultraviolet spectrophotometry at 450 nm

c.) Standard solutions preparations

- Prepare duplicate samples of blank and the standard solutions
- Treating in the same way as samples.

Calculation

Prepare a std. curve by plotting absorbance of std. against Manganese concentration.
Compute sample concentration from the curve.

D-Mn

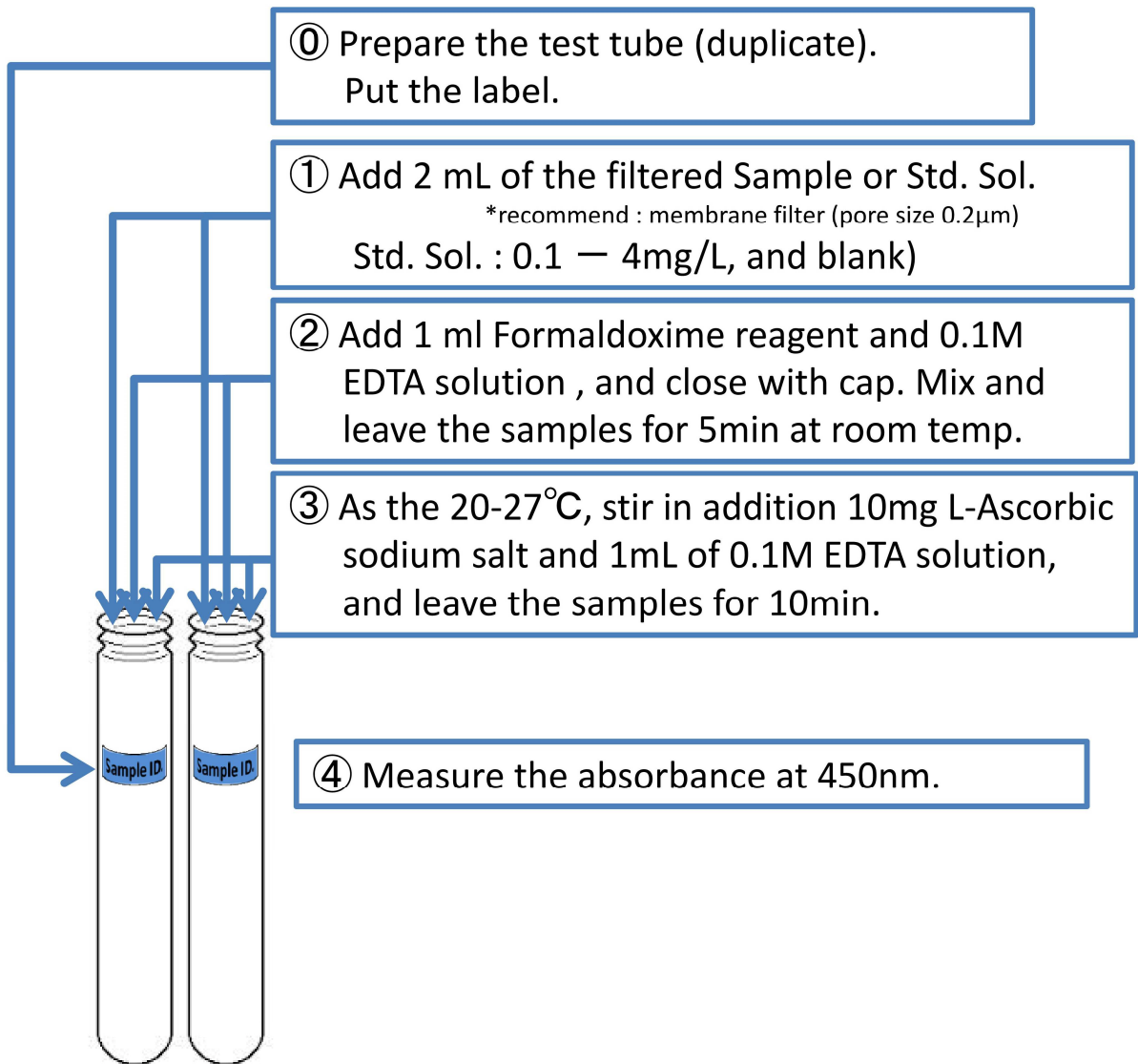


Fig. 12 Flow chart of the analytical procedure for D-Mn.

⑬ COD_{cr}(Closed Reflux) by Titrimetric Method (ref. : standard methods 22nd edition 5220 C)

Required Apparatus:

1. volumetric flask (1000ml, 500ml)
2. pipette (10ml, 5ml)
3. 200 ml measuring cylinder
4. bottle
5. 20 ml Ampule (clear) :
*Wash ampules with 20% H₂SO₄ before first use to prevent contamination.
6. Magnetic stirrer
7. Analytical balance
8. Burette
9. microburet
10. Autoclave

Interference:

Nitrite exerts a COD of 1.1mg O₂/mg NO₂⁻-N. Because concentration of NO₂⁻ in water rarely exceed 1 or 2 mg NO₂⁻-N/L, the interference is considered insignificant and usually is ignored. To eliminate a significant interference due to NO₂⁻, add 10mg Sulfamic acid for each mg NO₂⁻-N present in the sample volume used; add the same amount of Sulfamic acid to the ampoule containing the distilled water blank.

Required Chemicals:

1. Distilled water
2. conc. Sulfuric Acid : G.R.
3. 1.65 gm, Silver sulfate (Ag₂SO₄) : G.R.
4. 4.903 gm, Potassium dichromate (K₂Cr₂O₇) : primary standard grade
*Previously dried at 150°C for 2h.
5. 33.3 gm Mercuric sulfate (HgSO₄) : G.R.
6. 1.485 gm, 1, 10-Phenanthroline monohydrate : G.R.
7. 695 mg, FeSO₄.7H₂O : G.R.
8. 425 mg, Potassium Hydrogen Phthalate (KHP): G.R.
*Previously dried at 110°C.
9. 39.2 gm, Fe(NH₄)₂(SO₄)₂.6H₂O : G.R.
10. Sulfamic acid : G.R.

Preparation of reagents:

1. Sulfuric Acid reagent (Reagent A): (3.5ml/ampoule)
Dissolve 1.65 gm, Silver sulfate (Ag₂SO₄) in 0.3 kg of conc. Sulfuric Acid.
**Stir it for 1 day

2. Std. Potassium dichromate digestion solution (Reagent B) (0.01667 M): (1.5ml/ampoule)
To approximately 500 ml of distilled water, dissolve 4.903 gm of Potassium dichromate (99.98% pure), 167 ml of conc. H₂SO₄ and 33.3 gm of HgSO₄. Dissolve, cool to room temperature, and then dilute to 1000 ml.
****Take care when pouring Conc. H₂SO₄, pour slowly (not all at a time) in volumetric flask and stir gently to prevent overheating, keep in water bath with cold water**
3. Conc. Ferroin Indicator:
To approx. 50 ml of distilled water, dissolve 1.485 gm of 1,10-Phenanthroline monohydrate and 695 mg of FeSO₄·7H₂O and dilute to 100 ml.
4. Ferroin Indicator:
Take 10 ml of Conc. Ferroin indicator and dissolve to 40 ml distilled water.
5. Potassium Hydrogen Phthalate (KHP):COD_{Cr} value $\hat{=}$ 500mg-O₂/L
Dissolve 425 mg of KHP in 1000 ml of distilled water.
6. Standard Ferrous Ammonium Sulfate titrant (FAS) (0.10 M):
Dissolve 39.2 gm, Fe(NH₄)₂(SO₄)₂·6H₂O in distilled water. Add 20 ml of Conc. H₂SO₄ and cool and dilute to 1000 ml.
7. Ferrous Ammonium Sulfate titrant (FAS) (0.02 M):
200 ml of 0.10M FAS in distilled water make up to 1000mL.

Procedure:

- a. For Molarity of FAS solution:
 - i. Take 5 ml of Reagent B into 3 beakers each
 - ii. Add 10 ml of distilled water in all the beakers
 - iii. Put 2 drops of Ferroin indicator
 - iv. Titrate against FAS titrant

$$\text{Molarity of FAS solution (M)} = \frac{\text{Reagent B,ml}}{\text{Volume FAS used in titration,ml}} \times 0.1000$$

- b. Sample preparation: see Fig. 13
- i. triplicate all the samples
 - ii. place 2.5 ml of sample in ampule, add 1.5 ml of Reagent B. If high concentration of NO_2^- -N present, add 10mg Sulfamic acid for each mg NO_2^- -N present in the sample volume used
(Dilute the sample (if required) and take 2.5 ml of each)
 - iii. Carefully run 3.5 ml of Reagent A down inside the ampule so that an acid layer is formed under the sample digestion solution layer.
*** Do not mix.**
 - iv. The total volume in the ampule is 7.5 ml.
 - v. Take the ampules with samples and burn its upper part in order to seal it.
 - vi. After sealing, shake the ampules gently.
*** caution : wear face shield and protect hands from heat produced when contents of ampules are mixed. Mix thoroughly before applying heat to prevent local heating of ampule bottom and possible explosive reaction.**
 - vii. Check the solution color. Color is:
yellow : O.K.
green : Δ (If sample contents of SS or high DOC concentration, digestion reagent is not enough. Dilute a sample.)
blue : \times (digestion reagent is not enough. Dilute a sample.)
 - viii. Autoclave the ampules at 127°C for 2 h.
 - ix. After 2 hrs. of autoclave, cool to room temperature.
 - x. Open ampules, add 2 drops of Ferroin indicator and stir rapidly on magnetic stirrer while titrating with standardized 0.10M FAS.
 - xi. The end point is sharp color change from blue-green to reddish brown.
 - xii. Note down the volume of FAS used.
- c. Blank and check solution preparation:
triplicate blank and check solution
Blank : 2.5 ml of distilled water
Check solution : 1 ml of KHP solution, add 1.5ml of distilled water
Treat blank and std. solutions in same manner as samples.

Calculation:

$$\text{COD (mg-O}_2\text{/L)} = \frac{(A-B) \times M \times 8000}{\text{Sample volume ml}}$$

Where,

A= ml FAS used for blank,

B= ml FAS used for sample,

M= Molarity of FAS, and

8000= milliequivalent weight of oxygen $\times 1000$ mg/L

COD_{Cr}

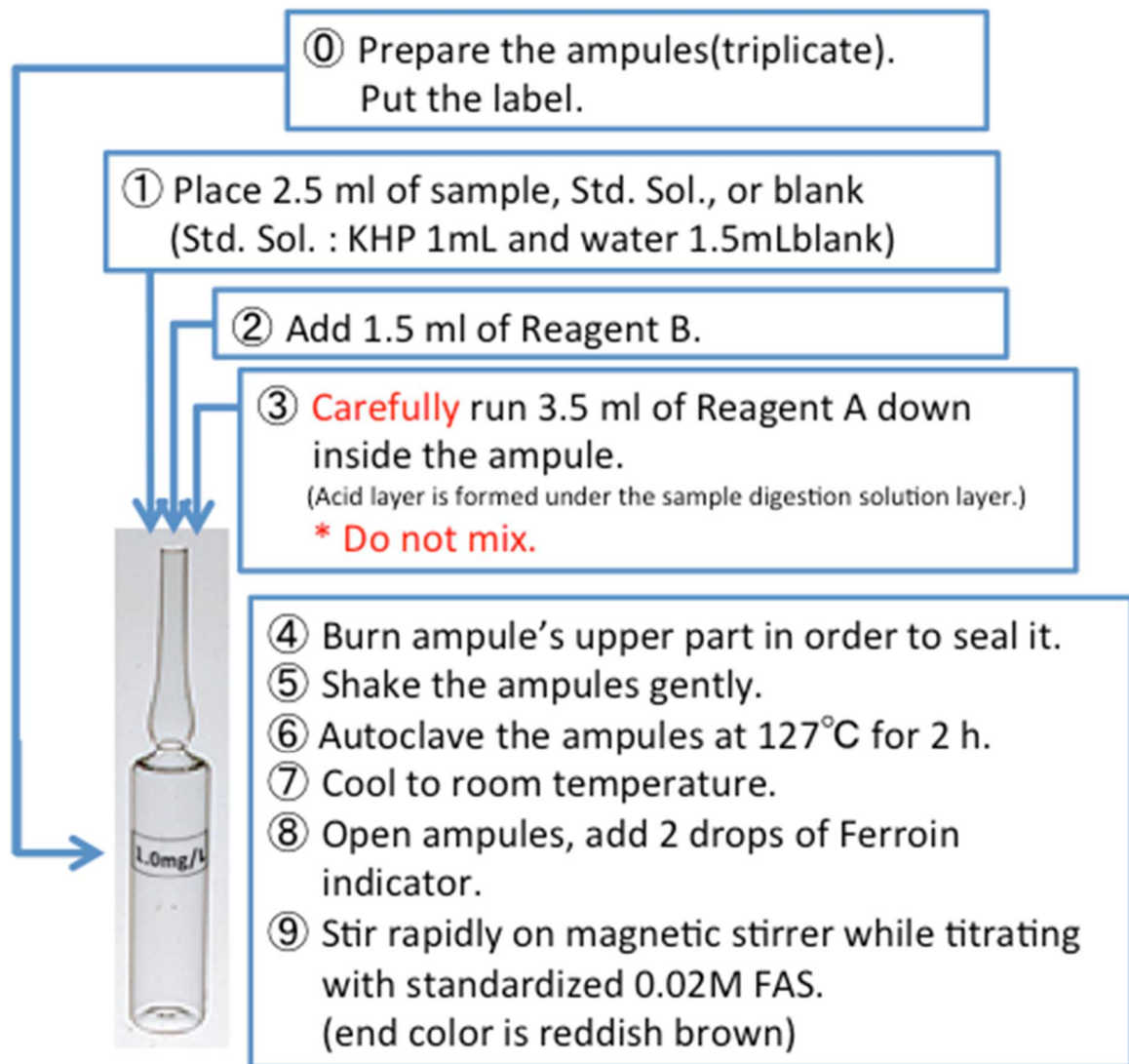


Fig. 13 Flow chart of the analytical procedure for COD_{Cr}.

⑭ Bicarbonate by Titration Method

(ref. : Standard Methods for the Examination of Tap Water in Japan)

Equipment:

- measuring cylinder (50mL, 100mL)
- dropper
- conical beaker(100mL)
- Burette

Apparatus:

- Magnetic stirrer
- Analytical balance

Required Chemicals:

9. Distilled water
10. 0.02N H₂SO₄ : for titration
11. Methyl Red : G.R.
12. Bromocresol Green : G.R.
13. Ethanol : G.R.

Preparation of reagents:

1. 95% Ethanol :
Mix 95mL Ethanol and 5mL water.
2. MR-BCG indicator : (5 drops/titration)
Dissolve 0.02 gm Methyl Red and 0.1gm Bromocresol Green, in 100 ml 95% Ethanol.
*** Store the indicator in plastic bottle and keep in refrigerator.**

Procedure(Titration): see Fig. 14

- vi. Measure 50mL sample into a 100mL conical beaker.
- vii. Add 5 drops of MR-BCG indicator and stir rapidly on magnetic stirrer while titrating with standardized 0.02N H₂SO₄ titrant.
- viii. The end point is sharp color change from blue (or green) to old pansy.
- ix. Note down the volume of 0.02N H₂SO₄ used.

Calculation

$$\text{HCO}_3^- (\text{mg/L}) = \frac{a(\text{mL})}{v(\text{mL})} \times 0.02(\text{N}) \times F \times 61(\text{g/mol}) \times 1000(\text{mg/g})$$

Where,

a= ml 0.02N H₂SO₄ titrant used,

v= ml sample volume,

F= factor of 0.02N H₂SO₄.

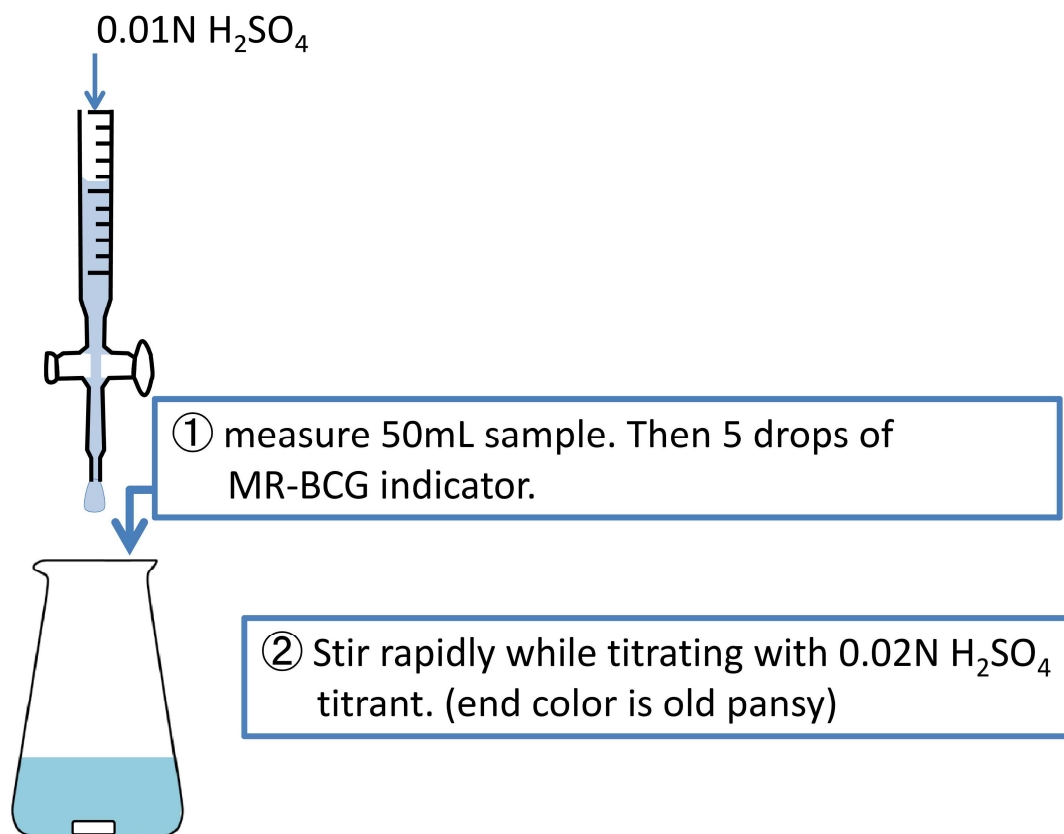


Fig. 14 Flow chart of the analytical procedure for HCO_3^- .

Wastewater treatment (in case of COD_{Cr})

The wastewater of COD_{Cr} method contains high concentration of acid, Mercury, and Chromium. Wastewater treatment process is necessary to throw safely away Drainage.

Water quality standard and Guidelines of those are as follows (table.1):

Table.1 Standard and Guidelines of water quality (mg/L)

		Drainage Standard (Japan)	Drinking water Standard (Japan)	Guidelines for Drinking water (WHO)
pH		5.8 < pH < 8.6		—
Hg		0.005	0.0005	0.001
Cr	Total	2	—	0.05
	Cr ⁶⁺	0.5	0.05	—

Equipment:

- Beaker
- Glass rod
- ORP meter
- pH test paper
- Filter
- Funnel
- Funnel stand
- Dropper
- Bottle
- PACKTEST (Cr⁶⁺, Cr·T, S)
- Mercury measurement set

Required Chemicals: **Low purity is enough**

1. Iron(II) Sulfate heptahydrate(FeSO₄ · 7H₂O)
2. Sodium Hydroxide (NaOH)
3. Calcium Hydroxide (Ca(OH)₂)
4. Sodium sulfide nonahydrate (Na₂S · 9H₂O)
5. Iron(III) Chloride hexahydrate (FeCl₃ · 6H₂O)
6. Hydrogen peroxide (30%, H₂O₂)

Preparation of reagents: in case of each 200ml wastewater treatment

1. 20% FeSO₄ Solution:
18g Iron(II) Sulfate heptahydrate in deionized water make up to 50ml
2. 10N NaOH Solution:
80g Sodium Hydroxide in deionized water make up to 200ml
3. 30% Ca(OH)₂ Slurry:
15g Calcium Hydroxide in deionized water make up to 50ml
4. 10% Na₂S Solution:
15g Sodium sulfide nonahydrate in deionized water make up to 50ml
5. 15% FeCl₃ Solution:
12.5g Iron(III) Chloride hexahydrate in deionized water make up to 50ml

Procedure: see p.4

- a. Reduction of Chromium (Cr⁶⁺ → Cr³⁺)
Measuring ORP and mixing by wastewater by glass rod while delivering 20% FeSO₄ Solution by drops into wastewater. When the ORP value decreases and the color change to blue-green (include Ferroin indicator) or yellow, stop the drop.
- b. Check the Chromium(VI) concentration
Measure Cr⁶⁺ concentration with Packtest (Cr₆₊; see p.5). If concentration exceeds the Drainage Standard of Japan, repeat a.
- c. Sedimentation (heavy metal)
***This process generates gases. Do the work outdoors or in Fume hoods.**
 - (1) Checking pH value by pH test paper and mixing by wastewater by glass rod while adding 10N NaOH. When the pH value is 7-8, stop the addition.
*** keep in water bath with cold water**
 - (2) Then, checking pH value by pH test paper and mixing by wastewater by glass rod while adding 30% Ca(OH)₂ Slurry. When the pH value is 9, stop the addition. Cool to room temperature.
- d. Filtration
Set up funnel, pleated filter paper (see p.6), beaker, and the funnel stand. Filter sediment from the liquid.

e. Check the Chromium concentration

Measure Cr concentration with Packtest (Cr • T; see p.7). If concentration exceeds the Drainage Standard of Japan, add a small amount of sulfuric acid, then back to c.

f. Sedimentation (Mercury)

***This process generates H₂S gas. Do the work outdoors or in Fume hoods.**

Measuring ORP and mixing by wastewater by glass rod while delivering 10% Na₂S Solution by drops into wastewater. When the ORP value is 0 or negative, stop the drop. Add a small volume of 15% FeCl₃ Solution and mix. Check the pH value (○ : pH > 9, × : pH < 9, add 10N NaOH). Let stand overnight.

g. Filtration

Set up funnel, pleated filter paper, beaker, and the funnel stand. Filter sediment from the liquid.

h. Check the Mercury concentration

First, check that the pH value of liquid is less than 9, and measure S²⁻ concentration (because, S²⁻ affects Hg measurement; see p.8). Measure Hg concentration with Mercury Measurement Set (see p.9-10). If concentration exceeds the Drainage Standard of Japan, back to f.

Sediment : After dry, store the sturdy bag

Liquid : colorless → drain

color → apply the liquid to sunlight until the color disappears, then drain

Wastewater Treatment

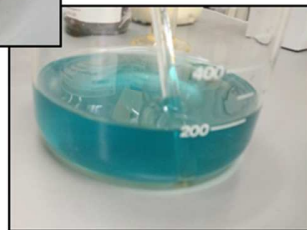
(in case of COD_{Cr})

a. Reduction

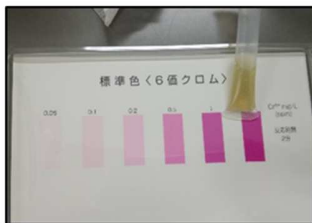


← initial

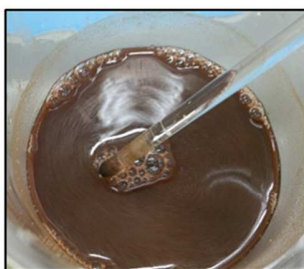
After reduction →



b. Concentration check (Cr^{6+})



c. Sedimentation



← mixing

After sedimentation →



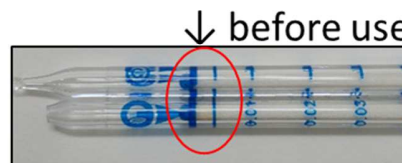
d. Filtration



f. Sedimentation (Hg)



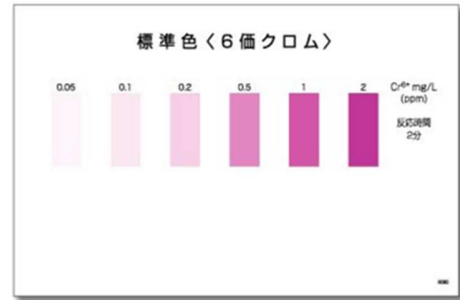
h. Concentration check (Hg)



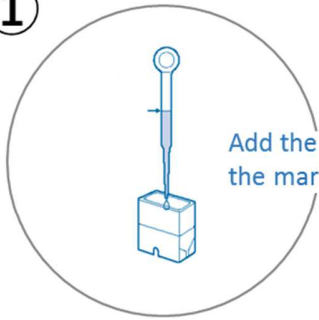
↓ before use

↑ after use

HOW TO USE . . . PACKTEST(Cr6+)

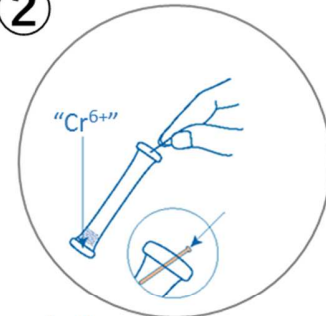


①



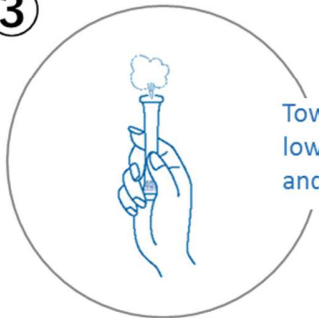
Add the sample with a pipette up to the mark

②



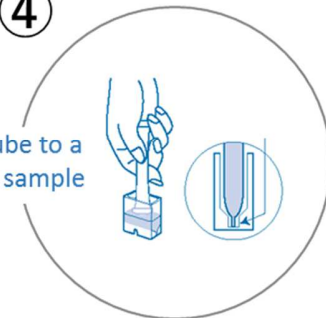
Pull out by pinching this part

③



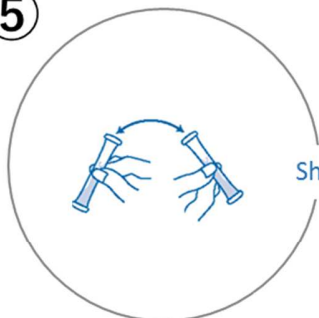
Toward a hole in the top, pinch strongly the lower half of the tube between the finger tips, and bent the tube into the V-shape

④



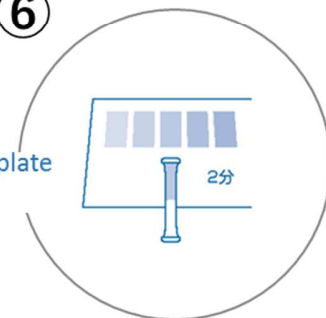
As it is put in the holes in the tube to a bottom of cup, inhale all of the sample

⑤



Shaking 5 times

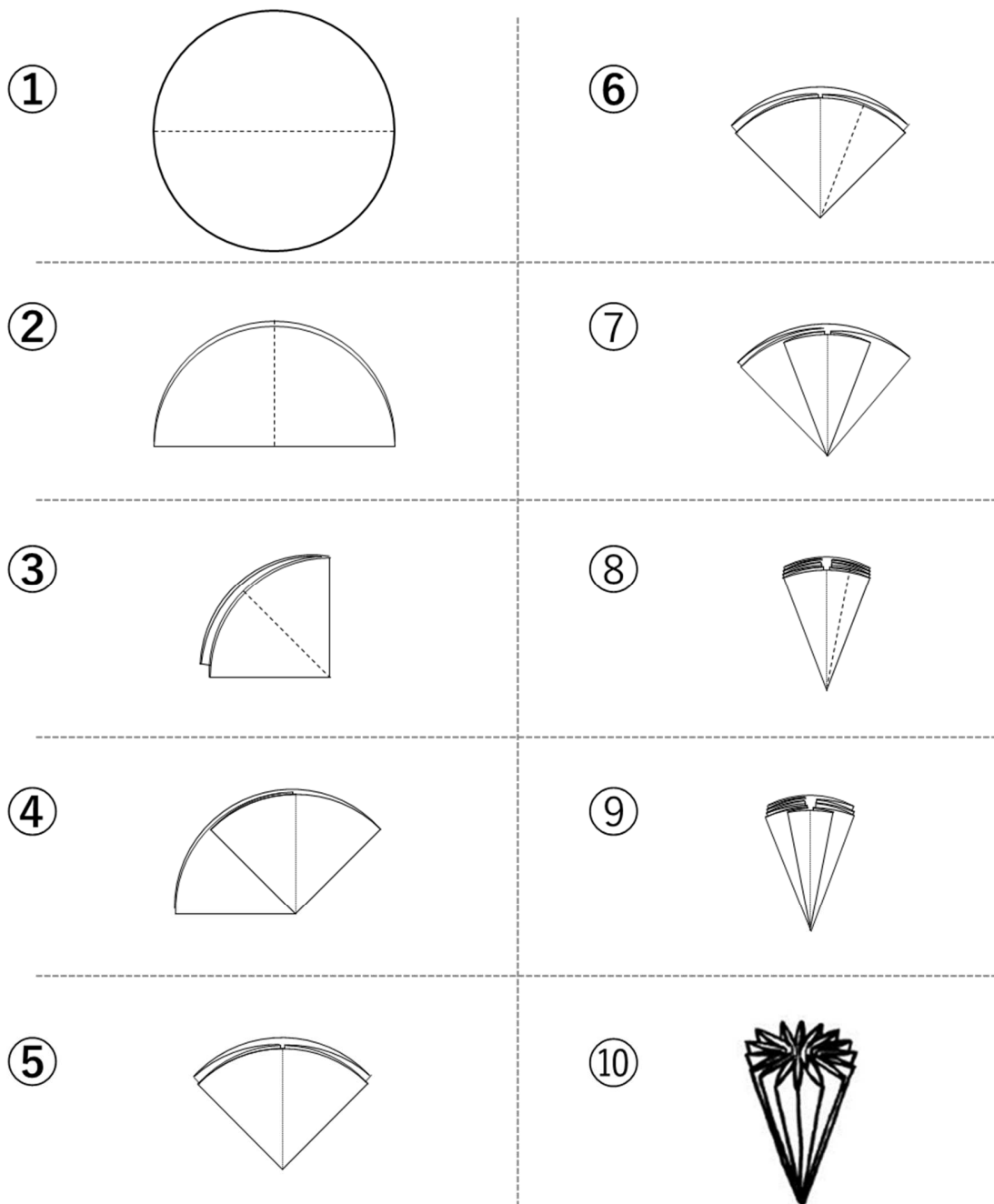
⑥



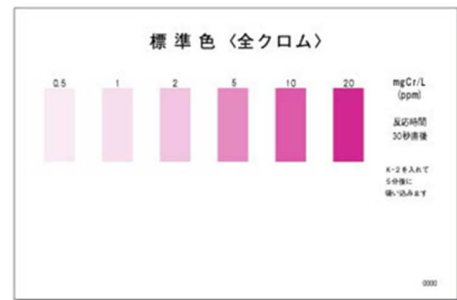
Compare the color of the color plate and tube after 2 minutes

How to make the pleated filter paper

As shown in the following figure, fold the filter paper many times.

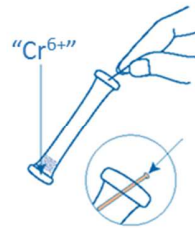


HOW TO USE・・・PACKTEST(Cr・T)



①

Add the sample with a pipette up to the mark (0.2mL)



⑥

Pull out by pinching this part



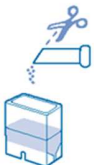
②

Add the K-1 solution with a pipette up to the mark (1.5mL)



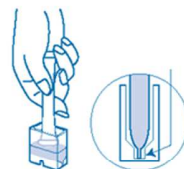
⑦

Toward a hole in the top, pinch strongly the lower half of the tube between the finger tips, and bent the tube into the V-shape



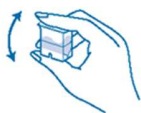
③

Add the K-2 powder



⑧

As it is put in the holes in the tube to a bottom of cup, inhale all of the sample



④

Close with the cap, and shake until the reagent has dissolved



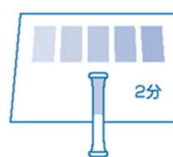
⑨

Shaking 5 times



⑤

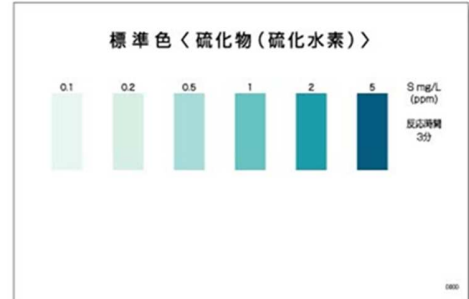
allow to stand for 5 minutes (Shake on the way once or twice)



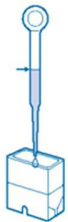
⑩

Compare the color of the color plate and tube after 30-second

HOW TO USE . . . PACKTEST(S)

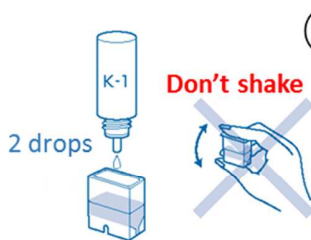


***Work of ①-⑤, be done quickly**



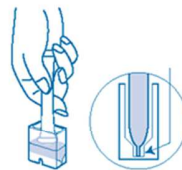
①

Add the K-1 solution with a pipette up to the mark (1.5mL)



②

Add the sample with a pipette up to the mark, and 2 drops pf k-1 solution



⑤

As it is put in the holes in the tube to a bottom of cup, inhale all of the sample



③

Pull out by pinching this part



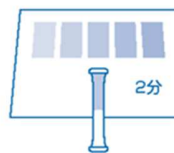
⑥

Shaking 5 times



④

Toward a hole in the top, pinch strongly the lower half of the tube between the finger tips, and bent the tube into the V-shape



⑦

Compare the color of the color plate and tube after 2 minutes

HOW TO USE

••• Mercury measurement set

1. Preparation

- ① Solution B : add a solvent for reagent B to the line and dissolve powder
(Adjusted at the time of use)
- ② 5M NaOH : 20g Sodium Hydroxide in deionized water make up to 100mL
- ③ Attach the connection rubber tube to the impinger (Fig.1)
- ④ Stand 2 tubes in a test tube stand, set 1ml of pipettes in each tubes (Fig.2)

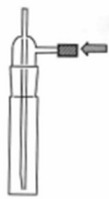


Fig.1 Impinger and the connection rubber tube

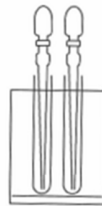


Fig.2 set tubes and pipettes

2. Measurement method

- ① check the pH value (○ : $1.0 < \text{pH} < 11.5$,
× : adjust sample so that a pH value becomes a range of 1 to 11.5)
- ② Transfer the 20ml sample to the impinger
- ③ Add 1mL Solution A, and 5mL 5M NaOH
- ④ Add 1mL Solution B
- ⑤ Break off the both ends of the detector tube and pretreatment tube
- ⑥ Set as follow(Fig.3):
Confirm the pump handle is fully pushed in. Then insert the detector tube into the rubber inlet with G ► mark towards the gas sampling pump.

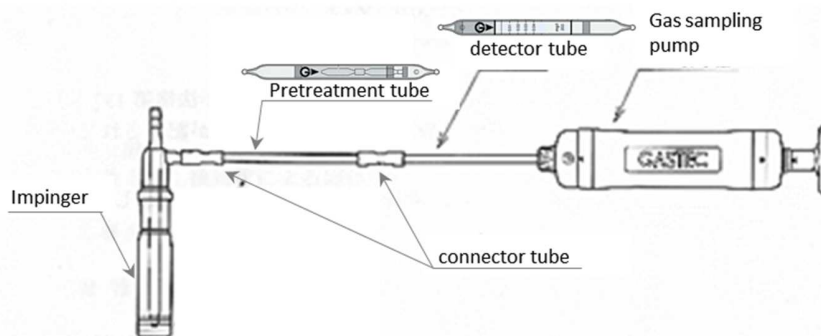


Fig.3 Connection of Mercury measurement kit

- ⑥ Align the guide marks on the pump shaft red (▲) 100 mL, and pull out the handle until it is locked. Wait until the sampling time has elapsed(1.5min).
- ⑦ Push back the handle, repeat ⑥ three times (total volume : 400mL)
- ⑧ After finish the absorption, remove the detector tube. The color in the detector tube changes as the gas is drawn in. Read the measurement at the end of the colored layer. (the color change : white - light orange)

3. Maintenance

Leakage of the gas sampling pump : (see table.1)

If you did not get rid of the bug, please refer to the repair request.

Table.1 The causes and solutions of leakage

	Causes	Solution
Inlet nut	The slack by having forgotten to close	Tighten
Inlet rubber	Damage or aging	Replace the part with new one
Cylinder, Piston	Pollution or lack of grease	Grease up, as follow:

- Maintenance of the gas sampling pump
 - (1) Give the tail block turns to the left, uncouple the piston from cylinder
 - (2) Clean the old grease inside of cylinder and outside of piston off with a soft or paper
 - (3) Apply grease the inside near the opening of cylinder
 - (4) Slide down the piston to the cylinder, give the tail block turns to the right, and screw that up tight
 - (5) Slide the piston in and out approximately 10 times, apply the grease to the cylinder
 - (6) do the air tightness test of the gas sampling pump, check the performance is good.

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