



Multidisciplinary study of trace metal biogeochemistry in the ocean

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1. Introduction

I am deeply honored to receive the 2022 Oceanochemistry Award. It has been my great pleasure to be able to establish close connections with many renowned Japanese chemical oceanographers through GEOTRACES program. Our collaboration have not only widely expanded my research scope but also have resulted in substantial research accomplishments. I would like to thank my Japanese friends for their open-handed support. Here, I would take this opportunity to introduce the studies and findings of my laboratory that we have applied multidisciplinary approaches and techniques to pursue scientific questions. I would also share some of the thoughts behind the questions and the processes I have switched topics from one to the other.

Going back to Taiwan in 2003 after my postdoctoral study at Princeton University, I first tried to measure intracellular trace metal quota in plankton assemblages that we collected in the adjacent oceanic regions. It turned out that the trace metal quota in size-fractionated plankton were much higher than what we have found in our culture study (Ho *et al.* 2003). I thus wondered where the elevated trace metals in or on the suspended particles are from in the oceanic regions around Taiwan, including the

Northern South China Sea, the Western Philippine Sea, and the Kuroshio water. To trace the sources of trace metals in the surface water, we had to develop analytical abilities to measure the elemental and isotopic composition of trace metals in seawater and particles. On the other hand, following the culture study learned at Princeton, I have been interested in studying trace metal requirement in some marine diazotrophs. At the same time, we have also applied laboratory culture technique and made the effort to establish biochemical and molecule biology techniques to study the roles of Ni in phytoplankton. Although the biochemical and molecular biology approaches we have used shall be pretty common in a molecular biology laboratory, the techniques are actually quite a challenge for a chemical oceanographer. However, I found that multidisciplinary approaches may provide convincing evidences to answer scientific questions proposed and thus to open interesting new questions.

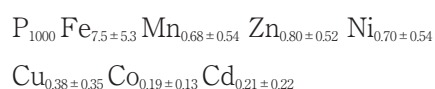
2. Trace metal quota in plankton assemblage

Can we extend the Redfield ratio to biologically active trace metals? This was the question we asked when we carried out the laboratory culture study to obtain the averaged

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elemental composition of marine phytoplankton about two decades ago when I was a postdoctoral fellow at Prof. François Morel's laboratory (Ho *et al.* 2003). The key concept of the Redfield ratio is not only the constant elemental composition (C, N, and P) in plankton assemblages collected from various oceanic regions. More importantly, the linkage and connection between the plankton composition in the surface water and major nutrient composition in the deep water establishes the internal biogeochemical cycling of the major elements and biomass in the global water column (Redfield 1934). The linkage of plankton biomass in the surface water and major nutrients in the deep water has served as a simple but critical foundation in marine biogeochemical studies. However, the biogeochemical cycling processes of trace metals should be much more complicated than major elements in the ocean.

By analyzing the elemental composition in 15 marine eukaryotic phytoplankton species representing 5 major eukaryotic phyla, we obtained the following P-normalized averaged elemental ratio from the 15 phytoplankton chosen (Ho *et al.* 2003; Ho 2006).



Although the quota of many trace metals obtained from the culture study seemed to be fairly close to the value obtained in the field, there were only a few field studies available reported reliable trace metal data two decades ago, which had to be justified by low Al content in the plankton or suspended particle samples. That was why measuring trace metal quota in field plankton samples was something I planned to do after going back to Taiwan. Luckily, Prof.

Liang-Saw Wen and Ms. Wen-Huei Lee from Institute of Oceanography National Taiwan University designed a trace metal clean size-fractionated plankton filtration collector at that time (Ho *et al.* 2007; Wen *et al.* 2018). Taking the advantage of the filtration apparatus, we have collected and measured trace metal composition in size-fractionated plankton in the oceanic regions around Taiwan for about 15 years. We found that the quota of many trace metals were one order of magnitude higher than the quota reported in the 2003 culture study (Ho *et al.* 2007; Liao *et al.* 2017). After 20 years, based on the highly varied plankton metal quotas observed in our and other scientists' field studies over all the global ocean, I would say that the surface-deep water Redfieldian linkage for major elements does not exist for most of the biologically active trace metals. The metal quota observed are highly varied spatially and are also significantly deviated from the dissolved metal to phosphate ratios observed in the deep water. In brief, as pointed out in the study of Ho *et al.* (2007), "The stoichiometry concept for trace-metal composition in marine plankton assemblages is unrealistic in the field".

First, although the concentrations of trace metals are relatively low in seawater and in biotic particles plankton intracellularly, they can be 'major' elements in abiotic particles (Ho *et al.* 2007; 2010a; 2011). For example, Fe and Al account for about 5 and 8 % of mass in lithogenic particles. Moreover, many trace metals are highly or partially particle reactive so that the dissolved form of the particle reactive metals tend to be removed or scavenged by other particles during transport processes, which explains why the dissolved concentrations of these metals exhibit a decreasing trend

vertically or horizontally through deep water circulation globally. In addition, operationally, it is highly challenged to well separate extracellularly aggregated and/or adsorbed particles with trace metals from plankton to determine intracellular metal quota. Although extracellular washing solution have been developed to remove extracellularly adsorbed or aggregated particles on plankton, the washing capability is based on complexation and redox reactions to solubilize the surface bound ions or amorphous precipitates. To the best of my knowledge, no systematic studies have been carried out to demonstrate that the washing solution is capable of removing lithogenic particles aggregated on biotic particles. Furthermore, intracellular trace metal composition can be highly varied due to the variations of ambient physical and chemical conditions, including the bioavailable concentrations of trace metals, the interactive relationship among different trace metals, the physiological conditions of phytoplankton due to varying abiotic conditions so that cellular trace

metal composition can be highly plastic (Figure 6 of Liao *et al.* 2017; Figure 7 of Zheng *et al.* 2021). Attributed to these complicated properties, almost all of biologically active trace metals do not show the Redfield type surface-deep water linkage.

However, probably being the only exception, Cd does exhibit the Redfieldian type distribution in the global ocean. Our early culture study reported that averaged intracellular Cd quota is $0.21 \pm 0.20 \text{ mmol mol}^{-1} \text{ P}$ (Ho *et al.* 2003). Cd to P ratios in size-fractionated plankton collected in the South China Sea showed similar ratios, ranging from 0.12 to 0.34 $\text{mmol mol}^{-1} \text{ P}$, and did not vary with Al and exhibited fairly consistent values among different sizes of plankton (Ho *et al.* 2009). In terms of dissolved concentrations, the dissolved Cd to phosphate ratios in the deep water slightly increases from 0.18 in the subarctic North Atlantic to about 0.33–0.35 mmol mol^{-1} in the northern Indian and Pacific Oceans (Baar *et al.* 1994). The most recent and comprehensive study in the Pacific Ocean shows that the dissolved Cd to P ratios at depth deeper

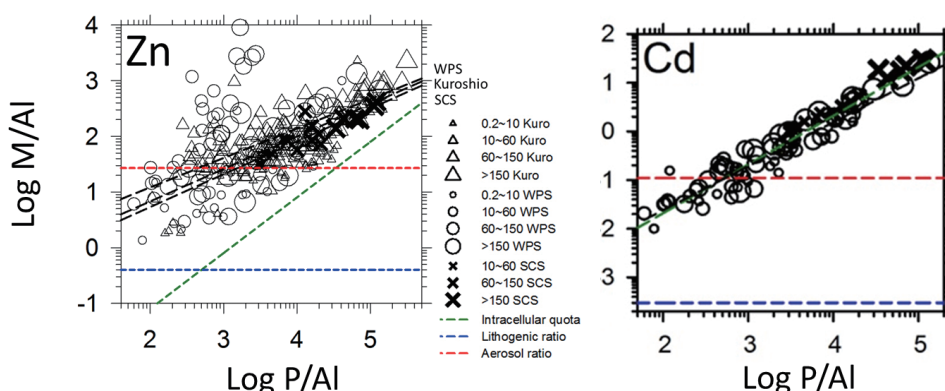


Fig 1. The elemental ratios, normalized to Al, in size-fractionated plankton or suspended particles collected in the South China Sea (SCS), the Western Philippine Sea (WPS), and the Kuroshio region (Kuro). The numbers shown next to the symbols are the size range (μm) for the plankton or particles collected in the three oceanic regions. We have used a simple mass balance model to trace the source of the metals, $[M]_i = a [Al]_a + b [Al]_l + c [P]_i$. The constants, a , b , c , stands for the elemental ratios (M/Al or M/P) in anthropogenic aerosols, lithogenic dusts, and cellular quota in plankton, respectively. The details may be found in the following references (Figure modified from the studies of Ho *et al.* 2007; Liao *et al.* 2017; Liao and Ho 2018).

than 1500 m are not only highly constant but also close to the plankton quota, which are 0.35 ± 0.02 , 0.32 ± 0.02 , and 0.35 ± 0.02 mmol mol⁻¹ P in the Southern Ocean, the South Pacific Ocean, and the North Pacific Ocean, respectively (Zheng *et al.*, 2021). Unlike Cd, the ratios of dissolved Zn to P ratios increase from the south to the north, which were 2.49 ± 0.40 , 3.12 ± 0.37 , and 3.70 ± 0.21 mmol mol⁻¹ P in the three regions from the south to the north, respectively (Zheng *et al.*, 2021; Liao *et al.* 2021).

In both suspended and sinking particles collected in the adjacent oceanic regions around Taiwan, we observed relatively high metal ratios in biotic particles (either normalized to P or Al), indicating the significant impacts of aerosol deposition on trace metal cycling in the oceanic regions (Ho *et al.* 2007; 2010a; 2011). By applying a simple mass balance approach, we found that anthropogenic aerosols are most likely to be the major source for the elevated metals. Using Zn as an example, the metal to P or Al ratios are most close to the composition in anthropogenic aerosols but not in lithogenic dusts or biotic particles (Fig. 1). These observations in both suspended and sinking particles have driven us to systematically study the impact of aerosol deposition on trace metal biogeochemistry in the oceanic regions.

3. Aerosol trace metal biogeochemistry: sources, internal transformation, and sinks

Aerosol deposition is known to be a dominant process for supplying trace metals to the surface water of the open ocean. As Figure 2 shows, the Northwestern Pacific Ocean (NWPO) and its marginal seas, right next to East Asia continent, receives tremendous amount of

both lithogenic and anthropogenic aerosols in the surface water (Fig. 2 and 3). Moreover, the seasonal exchange between the strong Northeastern and weak Southwestern monsoons results in dramatic seasonal fluctuations of aerosol deposition fluxes to the adjacent oceanic regions and thus provides us a natural platform in the region to study the transformation processes of aerosol trace metals in oceanic surface water between high and low deposition seasons. It appears that Taiwan occupies an excellent location to study aerosol trace metal biogeochemistry and to evaluate the impacts of aerosol deposition on marine phytoplankton growth and their community structure.

Although the elemental and isotopic composition of trace metals are powerful tracers and proxies to study physical and biogeochemical processes in the ocean, the sampling and analysis of the composition either in particulate or dissolved trace metals are highly challenging technically. It is essential to build the capacities of low contamination sampling and highly sensitive analytical techniques to accurately analyze trace metals at concentration levels ranging from nano- to picomolar in seawater. Before seawater pretreatment systems commercially available (e.g., *Seafast*, ESI), we had made the effort to custom-design fully automated flow injection analysis pretreatment systems to preconcentrate dissolved trace metal concentrations in seawater (Ho *et al.* 2010b; Wang *et al.* 2014). With the technical support of Japanese collaborators and colleagues from Academia Sinica, we have also gradually established analytical methods to measure the isotopic composition of some biologically active trace metals, including Cd, Zn, Fe, and Ni. We would like to take this

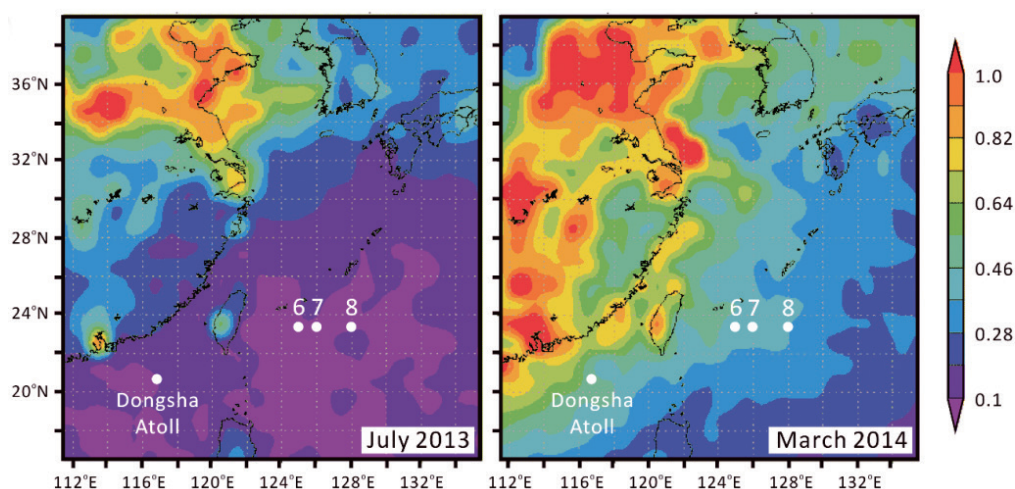


Fig 2. Monthly mean aerosol optical depth (AOD) in July 2013 and March 2014 acquired from MODIS-Aqua at 550 nm (<http://giovanni.gsfc.nasa.gov>). Field sampling stations 6, 7 and 8 in Taiwan GEOTRACES process study and Dongsha Atoll Research Station are marked (Figure modified from Wang and Ho 2020).

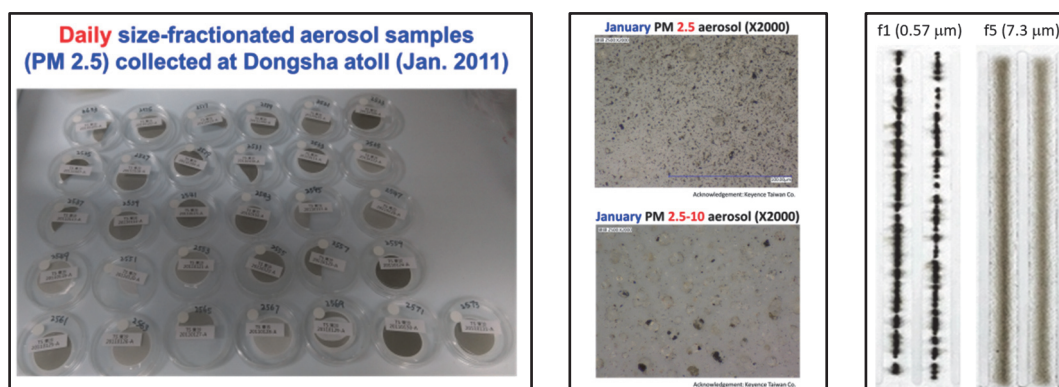


Fig 3. (Left) Daily size-fractionated aerosols, PM 2.5 and PM 2.5–10, collected at Dongsha atoll. (Middle) The images of 2000 fold magnification show that the size-fractionated aerosols were well separated. (Right) The smallest and largest size-fractionated aerosol samples collected in the East China Sea (Hsieh *et al.* 2022).

opportunity to thank Professors Yoshiki Sohrin, Shotaro Takano, Der-Chuen Lee, and Kuo-Fang Huang.

We first chose Cd to develop the elemental and isotopic study of trace metals in the ocean due to its highly constrained nutrient-type distribution in the global ocean and the relatively easy handling for contamination control during sampling and analysis. By measuring elemental and isotopic composition of Cd in seawater and particles, Mr. Shun-Chung Yang had carried out a series studies on Cd cycling in the NWPO and

its marginal seas (Yang *et al.* 2012; 2014; 2015; 2018). Being the first study targeting size-fractionated particulate samples, Yang *et al.* (2015) observed that Cd fractionation effects in the major biotic activities, phytoplankton uptake, microbial degradation and zooplankton repackaging, may take place at similar magnitudes and result in insignificant net biological fractionation effects in the surface water. Collaborating with the research groups of Prof. Jing Zhang and Yoshiki Sohrin during Yang's Ph.D. study period, we joined Japanese

GEOTRACES cruise KH-11-07, also known as GEOTRACES GP18, by R/V Hakuho-Marui in July 2011 to study Cd cycling in the Kuroshio-Oyashio extension region. The contrasting hydrographic distinction in the oceanic region provides us an ideal platform to investigate the spatial variations of Cd isotope fractionation systems. Yang *et al.* (2018) have studied the relative importance of physical and biogeochemical processes on Cd cycling by analyzing the distribution patterns of dissolved and particulate Cd isotopic composition in the region, demonstrating that Cd isotope fractionation can match either a closed or open system Rayleigh model, depending on the relative contribution of physical and biogeochemical processes on its cycling.

Attributed to the relatively low concentrations of Cd in aerosols and extremely high biological uptake rates by phytoplankton, the impacts of aerosol deposition are not easily observed for Cd in the ocean. We then switched to Zn cycling due to its extremely high concentrations and enrichment factors in aerosols and also its importance for phytoplankton growth. Liao *et al.* (2017; 2018) found that Zn and other trace metals exhibited elevated metal ratios (metal to P or Al) in size-fractionated plankton or suspended particles collected in the surface water of the Western Philippines Sea and the Kuroshio water. Liao *et al.* (2020) further investigated the distribution patterns of the elemental and isotopic composition of Zn in the water column of the NWPO and found that the isotopic composition in the deep water is lighter than the value observed in the Northeastern Pacific Ocean, indicating the influence of external sources in the oceanic region. We also observed that

aerosol optical depths are strongly associated with the variations of dissolved Zn elemental and isotopic composition in the surface water of the global ocean, supporting the argument that anthropogenic aerosol deposition plays an important role on causing the variations of the composition in oceanic surface water. In terms of the impacts in the deep water, Liao *et al.* (2021) found that anthropogenic aerosols are the major Zn source in the sinking particles collected by floating and moored sediment traps at SEATS, Taiwan's time series station in the South China Sea. These observations indicate that the deposition of anthropogenic aerosols plays a dominant role in regulating Zn elemental and isotopic composition in the surface water of large oceanic area and have even become a major source for Zn and some other trace metals in the deep water of some oceanic regions with high aerosol deposition fluxes (Takano *et al.* 2020).

During the past few years, we have started studies on aerosol Fe cycling in the ocean. It is well known that Fe supply to the ocean affects oceanic biological pump and carbon cycling globally. Aerosol deposition is a major process supplying external Fe to the surface ocean so that the quantification of aeolian bioavailable or dissolvable Fe supply in the surface water is significant in marine biogeochemistry. However, since the transformation of aerosol dissolvable Fe before and post-deposition in the surface water are involved in many complicated atmospheric and aquatic physicochemical processes and reactions, the quantification of the fluxes of aerosol dissolvable Fe in the euphotic zone has also been a very difficult task operationally. Both the deposition fluxes and the internal transformation processes of aerosol Fe

in oceanic surface water still largely remain unclear.

By comprehensively collecting and analyzing aerosols, seawater, size-fractionated suspended particles, and sinking particles during seasonally representative low and high aerosol deposition periods, the study of Wang and Ho (2020) had tried to investigate the transformation processes of aerosol Fe in the surface water of the Western Philippine Sea during high and low aerosol deposition seasons (Fig. 2). We found that biotic and abiotic suspended particulate Fe exhibited size-dependent distribution patterns. Lithogenic particles originating from aerosol deposition were mainly in particle fraction with sizes ranging from 2 to 25 μm in the surface water. Biotic particles smaller than 2 μm were the major carriers of intracellular and surface precipitated/aggregated Fe. Due to their dissimilar physical, chemical, and biological properties, these biotic and abiotic particles shall undergo different cycling processes and possess

significantly different Fe residence time and bioavailability in oceanic surface water (Fig. 4).

My Ph.D. student, Chih-Chiang Hsieh, has systematically studied the effects of aerosol sizes on the solubilities and the deposition fluxes of aerosol Fe in the East China Sea (Fig. 4). We found that the relative contribution of each size fraction on the masses of total, dissolved, and labile Fe varies dramatically. Due to the relatively high Fe solubilities but much lower deposition velocities for fine aerosols, we argued that most of the previous studies have significantly overestimated the deposition fluxes of aerosol dissolvable Fe (Hsieh *et al.* 2022). This study demonstrated the importance of carrying out size-fractionated aerosol sampling to accurately obtain the deposition fluxes of aerosol soluble Fe in the surface ocean. We have further observed element specific dissolution patterns for all other trace metals by using different leaching protocols (e.g., buffer or Berger leaches), reflecting the complicated and element specific

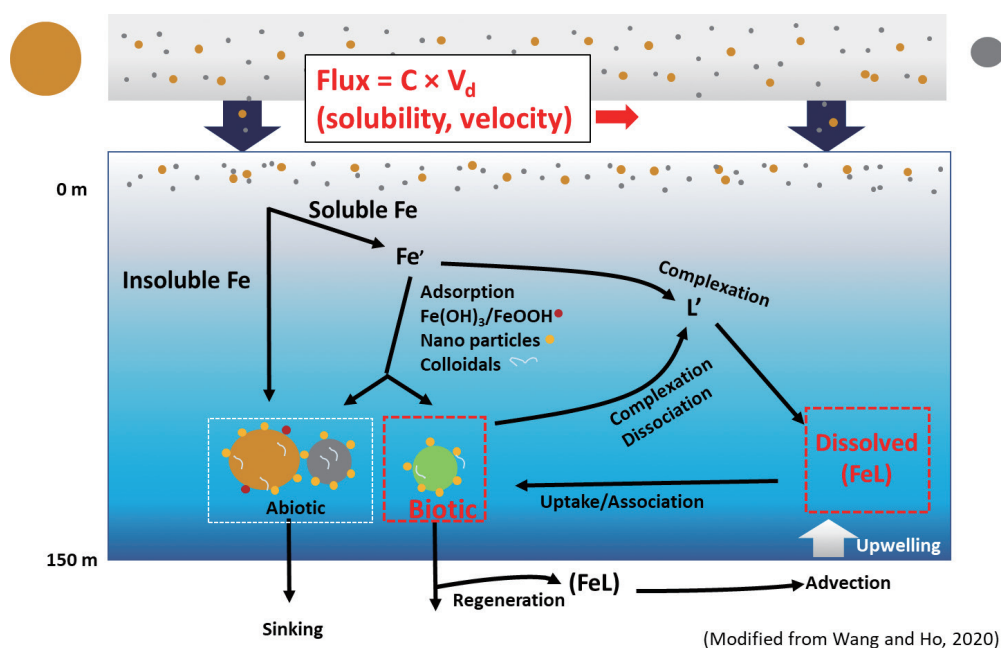


Fig 4. The schematic internal transformation of aerosol Fe in the surface ocean. The biotic inventories are composed of two portions: the intracellular and extracellular aggregated fractions (Figure modified from Wang and Ho (2020) by Chih-Chiang Hsieh).

effects of aerosol transport before and after aerosol deposition on the oceanic surface water (Hsieh *et al.* in prep.).

Our studies on Zn and Fe indicate that significant amount of soluble aerosol trace metals are transformed to labile particulate trace metals, which are most likely to be adsorbed and/or aggregated on small cyanobacteria in the surface water of the NWPO (Wang and Ho 2020; Liao *et al.* 2020, 2021; Hsieh *et al.* 2022). Trace metals in the labile particulate form are expected to hold relatively long residence time and high bioavailability in the euphotic zone. The internal transformation of the labile particulate trace metals originated from aerosol deposition remains to be explored.

4. The roles of Ni in diazotrophic phytoplankton

In addition to the geochemical approaches applied to study the elemental and isotopic composition in the dissolved and particulate field samples, we have also carried out laboratory phytoplankton culture experiments by using trace metal buffered culture techniques to investigate the roles of some trace metals in some specific marine phytoplankton or microalgae under specific growth conditions (Figs. 5–7). When an environmental variable shows promising evidence to be critical in marine biogeochemical studies, laboratory culture approaches are particularly useful to validate the hypothesis and the mechanisms proposed. The controlled physical and chemical variables may include light intensity, temperature, specific major or minor nutrients,

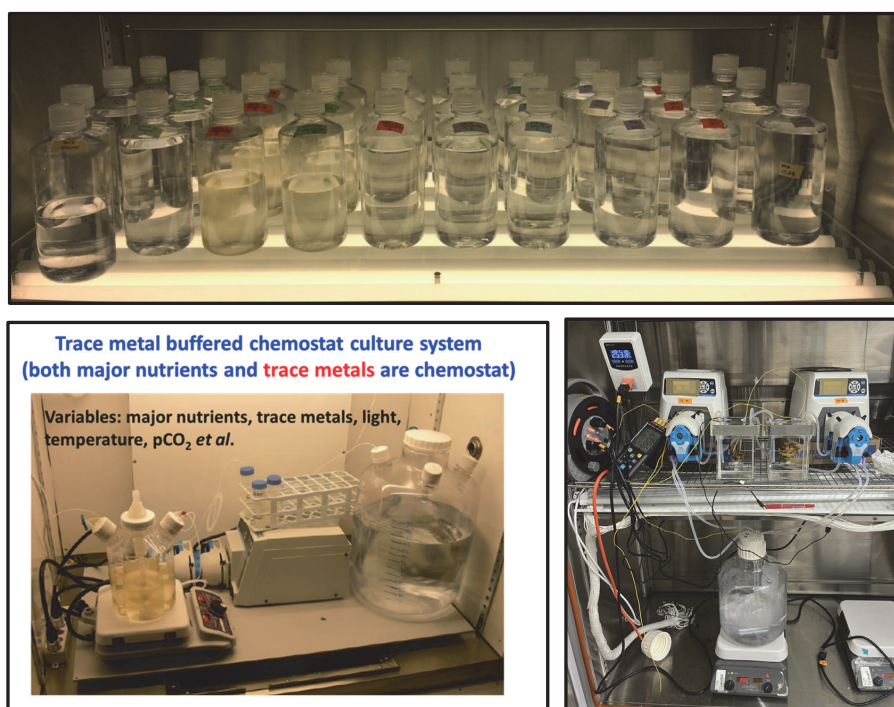


Fig 5. (Top) The trace metal buffered (by EDTA) batch phytoplankton culture experiments. (Bottom left) The chemostat culture system established in my laboratory. The source seawater may be mediated with various conditions as shown in the picture. (Bottom right) The chemostat culture system for coral bleaching experiments by varying seawater temperature, light intensities, and Fe availability (assembled by Melody Cheng and Chong-Jui Lee).

and the mutual interactions between physical and chemical parameters (Fig. 5). We have been particularly interested in studying the interaction between trace metals and marine diazotrophs simply because of the significance of nitrogen fixation in marine biogeochemistry.

Nitrogen (N_2) fixation is a critical process influencing carbon cycling in the ocean. However, our understanding on the major factors controlling oceanic N_2 fixation rates still largely remains unclear. Our interest on the demand of Ni in *Trichodesmium* was first attributed to the elevated Ni cellular quota in *Trichodesmium* observed both in the field and the laboratory culture. Moreover, the possession of Ni superoxide dismutase (NiSOD) and its unique capability to carry out photosynthesis and nitrogen fixation simultaneously have further driven our interest in pursuing the roles of Ni in *Trichodesmium*, a major N_2 fixation cyanobacterial group in the tropical and subtropical oceans (Fig. 6; Ho 2013; Chen *et al.* 2022). Without having heterocystous cells, *Trichodesmium* unusually carries out N_2 fixation and photosynthesis together. The diazotroph possesses the capability to bloom in the surface water where light intensities are extremely

strong. However, N_2 fixation is catalyzed by nitrogenase, an enzyme that is vulnerable to O_2 and oxidative stress generated in photosynthesis (Fig. 6). How *Trichodesmium* may cope with the oxidative stress to carry out nitrogen fixation under high light conditions is the question we would like to answer.

Applying trace metal defined culture techniques and biochemical approaches, we have gradually discovered the roles of Ni on N_2 fixation and H_2 production in some major marine diazotrophs (e.g., Ho *et al.* 2013; Tuo *et al.* 2020; Li *et al.* 2022; Chen *et al.* 2022). We have used custom-made antibodies of Ni-SOD and Ni-Fe hydrogenase and have applied them to investigate how Ni availability influences cyanobacterial N_2 fixation and H_2 production in the ocean (Fig. 6). Our culture studies have revealed that Ni availability is a critical factor on controlling H_2 production and N_2 fixation in some marine diazotrophic cyanobacteria (Tuo *et al.* 2020; Chen *et al.* 2022). We found that Ni is essential for *Trichodesmium* growth and can elevate its total superoxide dismutase (SOD) activities and N_2 fixation rates (Ho 2013). Sufficient Ni supply is essential for *Trichodesmium* to have high N_2 fixation rates

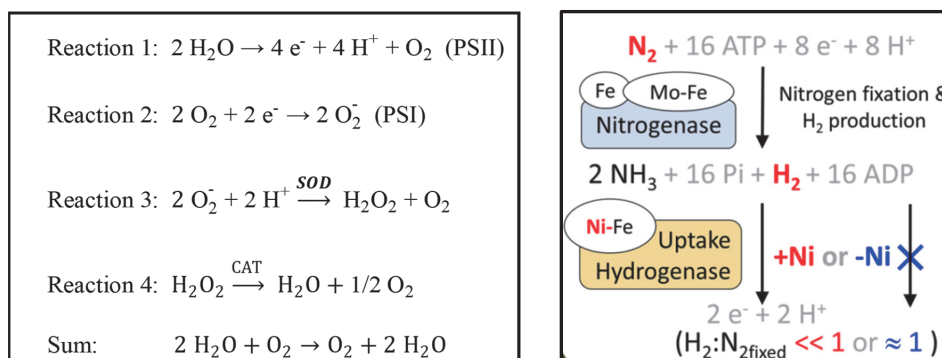


Fig 6. (Left) The importance of NiSOD in *Trichodesmium* through the Water-Water cycle in photosystems II and I (Asada 1999; Chen *et al.* 2022). (Right) By varying Ni availability, we observed the impacts of Ni on H_2 production through NiFe uptake hydrogenase in marine diazotrophic cyanobacteria (Tuo *et al.* 2020; Li *et al.* 2022).

under high light conditions (Ho *et al.* 2013; Rodriguez and Ho 2014). For the other diazotroph, *Cyanothece*, we found that Ni limitation results in one order of magnitude higher H₂ accumulation rates in the low Ni than high Ni treatments (Fig. 6, Tuo *et al.* 2020). Later on, we demonstrated that Ni deficiency decreases hydrogenase expression and leads to H₂ accumulation and N₂ fixation reduction in the marine diazotrophic cyanobacteria (Li *et al.* 2022).

Our recent study has further validated that NiSOD reduces oxidative stress generated during photosynthesis and thus protects its nitrogen fixation process under high light conditions (Chen *et al.* 2022). By producing and purifying the recombinant NiSOD protein through *E. coli*, we quantified NiSOD concentrations in *Trichodesmium* grown under various Ni and light treatments. The findings of the study may explain why *Trichodesmium* can carry out N₂ fixation and photosynthesis simultaneously and survive and bloom in the surface water of the tropical ocean. These results may also shed light on the mechanisms of photoinhibition for phytoplankton under high solar radiation conditions (Chen *et al.* 2022).

Biological Ni demand may vary significantly

among different diazotrophic cyanobacterial groups, attributed to the difference of their intracellular Ni-containing enzymes and requirement. Both *Trichodesmium* and *Crocospaera* contain Ni-Fe hydrogenase, Ni-SOD, and urease. For future study, we would investigate the Ni requirement of various diazotrophic cyanobacterial groups, the expressions of Ni-containing enzymes, and the impacts of Ni availability on N₂ fixation and H₂ production under different growth conditions. We are currently applying our custom-made antibodies and purified Ni-SOD and Ni/Fe hydrogenases to quantify the enzymatic concentrations in *Trichodesmium*, *Cyanothece*, and other diazotrophic cyanobacteria both in laboratory culture and the field. *In situ* Ni enrichment experiments would be carried out to validate whether natural Ni concentrations are sufficient or not for different diazotrophs to reach their maximum nitrogen fixation rates under conditions with various light intensities and temperature gradients (Fig. 7).

In addition to *Trichodesmium*, we have extended the concept of reactive oxygen species-trace metal demand to endosymbiotic dinoflagellates (or *Symbiodinium*). Most of global coral reefs have been facing serious bleaching



Fig 7. (Left) We carried out field experiments in the Kuroshio water to validate the importance of Ni availability on the nitrogen fixation rates in *Trichodesmium* under high light conditions. (Right) The colony of *Trichodesmium* picked up from the surface water of the Kuroshio.

threats and the bleaching problem has been mainly attributed to thermal stress and oxidative stress. We have first systematically studied trace metal requirement in a model *Symbiodinium*. The systematic studies have exhibited the fundamental understanding for trace metal requirement in endosymbiotic dinoflagellates (Rodriguez *et al.* 2016; Rodriguez and Ho 2017, 2018; Reich *et al.* 2020; Reich *et al.* 2021). We found that *Symbiodinium* possesses high Fe and Zn requirement. Our preliminary studies show that Fe requirement in *Symbiodinium* is significantly elevated at relatively high ambient seawater temperature (Reich *et al.* 2021). We are currently carrying out culture experiments with hard corals to demonstrate whether Fe availability is a critical factor affecting coral bleaching (Fig. 5). Hopefully we may share exciting findings to you in the near future.

5. Final Remarks

Trace metal supply to oceanic surface water is a major factor controlling oceanic primary production, phytoplankton community structure, nitrogen fixation process, and carbon cycling. In spite of their important roles on material cycling and biogeochemical processes in the ocean, the controlling processes and mechanisms of marine trace metal cycling still largely remains to be explored. Marine trace metal biogeochemistry itself is actually a multidisciplinary field. It seems to me that biochemical approaches are critical or even unavoidable for many key marine biogeochemical studies. However, many marine biogeochemists may not equip with the analytical tools in both geochemistry and biochemistry in their laboratories. Based on my personal experiences, I feel that the effort spent

to build up the multidisciplinary capacities is highly rewarded for marine trace metal biogeochemical studies.

6. Acknowledgements

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[References]

- Asada, K. 1999. The water-water cycle in chloroplasts: scavenging of active oxygens and dissipation of excess photons. *Annual review of plant biology* 50: 601–639.
- Chen, C.-C., I. B. Rodriguez, Y.-l. L. Chen, J. P. Zehr, Y.-R. Chen, S.-T. D. Hsu, S.-C. Yang, and T.-Y. Ho (2022) Nickel superoxide dismutase protects nitrogen fixation in *Trichodesmium*. *Limnology and Oceanography Letters* doi: 10.1002/lol2.10263.
- Baar, H. J. W. D., Saager, P. M., Nolting, R. F., & Meer, J. V. D. (1994). Cadmium versus phosphate in the world ocean. *Marine Chemistry*, 46(3), 261–281.
- Ho, T.-Y., A. Quigg, Z. V. Finkel, A. J. Milligan, K. Wyman, P. G. Falkowski, and F. M. M. Morel (2003) The elemental composition of some marine phytoplankton. *Journal of Phycology* 39: 1145–1159.
- Ho, T.-Y. (2006) The trace metal composition of marine microalgae in cultures and natural assemblages. pp 271–299. In *Algal Cultures, Analogues of blooms and Applications*.

Edited by DV Subba Rao, Science Publishers, New Hampshire, USA.

- Ho, T.-Y., L.-S. Wen, C.-F. You, and D.-C. Lee (2007) The trace metal composition of size-fractionated plankton in the South China Sea: biotic versus abiotic sources. *Limnology and Oceanography* 52: 1776–1788.
- Ho, T.-Y., C.-F. You, W.-C. Chou, S.-C. Pai, L.-S. Wen, and D. D. Sheu (2009) Cadmium and phosphorus cycling in the water column of the South China Sea: the roles of biotic and abiotic particles. *Marine Chemistry* 115: 125–133.
- Ho, T.-Y., W.-C. Chou, C.-L. Wei, F.-J. Lin, G. T. F. Wong, and H.-L. Lin (2010a) Trace metal cycling in the surface water of the South China Sea: Vertical fluxes, composition, and sources. *Limnology and Oceanography* 55: 1807–1820.
- Ho, T.-Y., C.-T. Chien, B.-N. Wang, and A. Siriraks (2010b) Determination of trace metals in seawater by an automated flow injection ion chromatograph pretreatment system with ICPMS. *Talanta* 82: 1478–1484.
- Ho, T.-Y., W.-C. Chou, H.-L. Lin, and D. D. Sheu (2011) Trace metal cycling in the deep water of the South China Sea: The composition, sources, and fluxes of sinking particles. *Limnology and Oceanography* 56: 1225–1243.
- Ho, T.-Y. (2013) Nickel limitation of nitrogen fixation in *Trichodesmium*. *Limnology and Oceanography* 58: 112–120.
- Ho, T.-Y., T.-H. Chu, and C.-L. Hu (2013) Interrelated influence of light and Ni on *Trichodesmium* growth. *Frontiers in Microbiology* doi: 10.3389/fmicb.2013.00139.
- Hsieh, C.-C., H.-Y. Chen, and T.-Y. Ho (2022) The effect of aerosol size on Fe solubility and deposition flux: A case study in the East China Sea. *Marine Chemistry* doi: 10.1016/j.marchem.2022.104106.
- Liao, W.-H., S.-C. Yang, and T.-Y. Ho (2017) Trace metal composition of size-fractionated plankton in the Western Philippine Sea: The impact of anthropogenic aerosol deposition. *Limnology and Oceanography* 52: 2243–2259.
- Liao, W.-H. and T.-Y. Ho (2018) Particulate trace metal composition and sources in the Kuroshio adjacent to the East China Sea: the importance of aerosol deposition. *Journal of Geophysical Research: Oceans* doi: 10.1029/2018JC014113.
- Liao, W.-H., S. Takano, S.-C. Yang, K.-F. Huang, Y. Sohrin, and T.-Y. Ho (2020) Zn isotopic composition in the water column of the Northwestern Pacific Ocean: the importance of external sources. *Global Biogeochemical Cycles* doi: 10.1029/2019GB006379.
- Liao, W.-H., S. Takano, H.-A. Tian, H.-Y. Chen, Y. Sohrin, and T.-Y. Ho (2021) Zn elemental and isotopic features in the sinking particles of the South China Sea: the implications to its sources and sinks. *Geochimica et Cosmochimica Acta* doi: 10.1016/j.gca.2021.09.013.
- Li, H.-T., S. Tuo, M.-C. Lu, and T.-Y. Ho (2022) The effects of Ni availability on H₂ production and N₂ fixation in a model unicellular diazotroph: The expression of hydrogenase and nitrogenase. *Limnology and Oceanography* doi: 10.1002/lno.12151.
- Redfield, A. C. 1934. On the proportions of organic derivatives in sea water and their relation to the composition of plankton. In Daniel, R. J. [Ed.] James Johnstone Memorial Volume. Liverpool University Press, pp. 176–92.

- Reich, H. G., I. B. Rodriguez, T. C. LaJeunesse, and T.-Y. Ho (2020) Endosymbiotic dinoflagellates pump iron: differences in iron and other trace metal needs among the Symbiodiniaceae. *Coral Reefs* doi: 10.1007/s00338-020-01911-z.
- Reich, H. G., W.-C. Tu, I. B. Rodriguez, Y. Chou, E. F. Keister, D. W. Temp, T. C. LaJeunesse, and T.-Y. Ho (2021) Iron availability modulates the response of endosymbiotic dinoflagellates to heat stress. *Journal of Phycology* doi: 10.1111/jpy.13078.
- Rodriguez, I. B. and T.-Y. Ho (2014) Diel nitrogen fixation pattern of *Trichodesmium*: the interactive control of light and Ni. *Scientific Reports* doi: 10.1038/srep04445.
- Rodriguez I. B., S. Lin, J. Ho, and T.-Y. Ho (2016) Effects of trace metal concentrations on the growth of the coral endosymbiont *Symbiodinium kawagutii*. *Frontiers in Microbiology* doi: 10.3389/fmicb.2016.00082.
- Rodriguez, I. B. and T.-Y. Ho (2017) Interactive effects of spectral quality and trace metal availability on the growth of *Trichodesmium* and *Symbiodinium*. *PLoS ONE* doi: 10.1371/journal.pone.0188777.
- Rodriguez, I. B. and T.-Y. Ho (2018) Trace metal requirements and interactions in *Symbiodinium kawagutii*. *Frontiers in Microbiology* doi: 10.3389/fmicb.2018.00142.
- Takano, S., W.-H. Liao, H.-A. Tian, K.-F. Huang, T.-Y. Ho, and Y. Sohrin (2020) Sources of particulate Ni and Cu in the water column of the northern South China Sea: evidence from elemental and isotope ratios in aerosols and sinking particles. *Marine Chemistry* doi: 10.1016/j.marchem.2020.103751.
- Tuo, S., I. B. Rodriguez, and T.-Y. Ho (2020) H₂ accumulation and N₂ fixation reduction by Ni limitation in *Cyanothece*. *Limnology and Oceanography* doi: 10.1002/lno.11305.
- Wang, B.-S., C.-P. Lee, and T.-Y. Ho (2014) Trace metal determination in natural waters by automated solid phase extraction system and ICP-MS: the influence of low level Mg and Ca. *Talanta* 128: 337-344.
- Wang, B. - S. and T. - Y. Ho (2020) Aerosol Fe cycling in the surface water of the Northwestern Pacific Ocean. *Progress in Oceanography* doi: 10.1016/j.pocean.2020.102291.
- Wen, L.-S., Lee, C.-P., Lee, W.-H., & Chuang, A. (2018). An ultra-clean multilayer apparatus for collecting size fractionated marine plankton and suspended particles. *Journal of Visualized Experiments*, 134(134), e56811. <https://doi.org/10.3791/56811>
- Yang, S.-C., D.-C. Lee, and T.-Y. Ho (2012) The isotopic composition of Cadmium in the water column of the South China Sea. *Geochimica et Cosmochimica Acta* 98: 66-77.
- Yang, S.-C., D.-C. Lee, T.-Y. Ho, L.-S. Wen, and H.-H. Yang (2014) The isotopic composition of dissolved cadmium in the water column of the West Philippine Sea. *Frontiers in Marine Science* doi: 10.3389/fmars.2014.00061.
- Yang, S.-C., D.-C. Lee, and T.-Y. Ho (2015) Cd isotopic composition in the suspended and sinking particles of the surface water of the South China Sea: the effects of biotic activities. *Earth and Planetary Science Letters* 428: 63-72.
- Yang, S.-C., J. Zhang, Y. Sohrin, and T.-Y. Ho (2018) Cadmium cycling in the water column of the Kuroshio-Oyashio Extension region: Insights from dissolved and particulate isotopic composition. *Geochimica et Cosmochimica Acta* doi: 10.1016/j.

gca.2018.05.001.

Zheng, L., T. Minami, S. Takano, T.-Y. Ho, and Y. Sohrin (2021) Sectional distribution patterns of Cd, Ni, Zn, and Cu in the North Pacific

Ocean: systematic importance of scavenging. *Global Biogeochemical Cycles* doi: 10.1029/2020GB006558.