
Jianyin Huang

A thesis submitted in fulfilment of the requirements of the degree of

Doctor of Philosophy

Environmental Future Research Institute
Griffith School of Environment,
Griffith University, Australia

November 2015
Acknowledgements

Thanks must firstly go to my supervisors: Professor Peter Teasdale, Associate Professor David Welsh, and Dr William Bennett. I am very grateful to have been your student and for all you help with the problems and difficulties of my PhD study. Pete, you have been very supportive for my study and gave me many opportunities. You managed to read my manuscripts although you were under a lot of pressure from your teaching. Dave, you were the one that I annoyed the most but you still answered my questions patiently and gave me so many ideas about the experiments. Thanks for giving me a big hug when I needed support. Will, you always knew that I was going to ask you questions or for help when I looked at you so happily. Thank you so much for all your support, encouragement and guidance!

Thanks to Ryan, Jeremy and Allen for letting me use the nutrient analyser. Thanks Amir, Nadeeka, Charlene, Noemie, Josh, Mohammad, Tao, and Sheng for supporting my PhD study. Thanks Michael, even though I still could not understand your bad jokes. I am glad that you treated me like family and supported me no matter what. Special thanks go to Tianling, Fan and Sean for being my volunteers and helping me so much in the field. Without you, I would not have managed to do all the field experiments.

Thanks to Griffith School of Environment for my PhD scholarship and completion scholarship, and for funding my research.

Finally, I would like to thank my family. Far away from my family for four-year study in Australia made me understand how important you are to me. I think you will be proud of me, but I am also proud to have you as my parents and grandparents.
Statement of Originality

The material presented in this report has not been previously submitted in any University and, to the best of my knowledge, contains no material previously published or written by another person except where due acknowledgement is made in the report itself.

Jianyin Huang

November 2015
Abstract

Due to rapid industrialisation, the growing global population, and the impacts of climate change, the availability and quality of water resources around the world has become degraded. Some regions are suffering water shortages despite sufficient water reserves, because surface and ground waters are contaminated to such a degree that they have become inadequate for potable use. Agricultural practices, industrial discharges and human wastewater are responsible for most contamination of water.

Nitrogen (ammonium, nitrate and nitrite) and phosphorus (phosphate) are the most common nutrients in freshwaters and estuaries that impact water quality. Excess nutrient loadings to water bodies can affect many aquatic organisms and, ultimately, contribute to the degradation of freshwater, estuarine and coastal marine ecosystems. Furthermore, high concentrations of nutrients in drinking water sources can cause health impacts to human beings. Due to the potential sporadic nature of the contamination sources, grab sampling may fail to identify contamination events. Nutrient loadings to waterways from point and non-point sources are of major ecological concern and represent one of the most significant water quality issues in surface water bodies, and hence require use of accurate and representative approaches to monitor nutrient concentrations. DGT as diffusive gradients in thin films, a well-established passive sampling technique, allows determination of time-weighted average measurements over environmentally relevant time-scales.

In this thesis, DGT techniques were developed for nitrate, for the first time, and evaluated using commercially-available anion exchange materials – Purolite A520E resin and AMI-7001 membrane which was used directly as a DGT binding layer. New DGT techniques are also described and evaluated for ammonium using cation exchange materials – Microlite
PrCH resin and CMI-7000 membrane. Performance characteristics were very promising and likely suitable for deployment in many freshwaters, especially those with conductivity < 1000 µS cm\(^{-1}\). These new DGT techniques were applied in different field sites to investigate their performance and to allow comparison with concentrations obtained from frequent grab sample collection and analysis. The in situ DGT techniques provided accurate time-weighted average concentrations that were highly representative of the inorganic nutrient concentrations for short-term (24 h) and long-term (72 h) deployments and were complementary to other measurements. These new DGT techniques should therefore prove to be a highly useful and inexpensive tool for use in comprehensive nutrient monitoring programs.
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1. Introduction

1.1 Urban wastewater

The continued existence and development of cities relies on the availability of good quality water. Urban water supply is a basic factor for economic development and public health. In the less-developed areas of the world, water-borne diseases such as cholera, typhoid and paratyphoid fevers, amoebic dysentery and diarrhoeal disease are common and well acknowledged.\(^1\)\(^-\)\(^3\) The pollution of water resources by human waste is the chief reason for the spread of enteric diseases.\(^1\) As populations and urbanisation expand, reclaimed water is becoming an increasingly important water resource because of the shortage of freshwater in urban areas.\(^4\)\(^-\)\(^6\) Reclaimed water has been used in urban areas for toilet flushing, water channel restoration and plant irrigation.\(^4\)\(^,\)\(^6\) The quality of the reclaimed water relies on the efficiency of wastewater treatment and can be affected by toxicants in the source waters.\(^4\)\(^,\)\(^5\)\(^,\)\(^7\) The growth of urban areas causes significant changes in local hydrological properties, generating urban runoff that reaches water bodies, faster and with greater force, leading to the degradation of ecosystems.\(^8\) There are strong corelations between urbanisation, stormwater and wastewater discharges, the transfer of pollutants, and the quality of natural surface and ground waters.\(^8\) The main sources of urban water contaminants – stormwater, wastewater from domestic, commercial and industrial activities, and effluents from wastewater treatment plants are presented below.

In urban areas, rain that falls on man-made impermeable surfaces such as building roofes, paved areas, footpaths, roads and driveways is collected and carried away through the urban catchment of stormwater drains that is usually separate to the sewerage system. Urban stormwater contains dissolved, colloidal and solid constituents in a heterogeneous mixture which includes nutrients, heavy metals, oils, greases, organic and inorganic compounds.\(^9\)
Stormwater Quality Improvement Devices (SQIDs) have been utilised to trap or collect rubbish and pollution that end up in stormwater drains, which can prevent large quantities of pollutants entering the stormwater drainage system.\textsuperscript{10} However, SQIDs are not effective in removing all contaminants, for instance, soluble nutrients, heavy metals, organics, suspended solids and salts can still flow from streets and gutters into creeks, streams, rivers, harbours and the ocean.\textsuperscript{11,12}

Urban wastewater is mainly from domestic, commercial and light industrial sources. Domestic inputs originate predominantly from human waste, and various household and office activities. Light industrial estate inputs include a number of automotive-related practices, electrical wholesalers, cleaning contractors, glazing and office supplies/printing. Industrial inputs that are discharged to the sewage system normally have large volumes, high concentrations and specific point sources.\textsuperscript{13} Urban wastewater generally contains large amounts of nutrients,\textsuperscript{14} heavy metals,\textsuperscript{15} salts and priority pollutants.\textsuperscript{16-18}

Wastewater treatment plants (WWTPs), including municipal and industrial districts, receive complex mixtures of pollutants and reduce their concentrations to reduce the impacts on the receiving environment.\textsuperscript{19} Many contaminants are removed or partially removed by WWTPs, however, some of them, such as nutrients, remain in substantial quantities.\textsuperscript{20} About 90\% of nutrient loads can be removed during wastewater treatment.\textsuperscript{21} Total nitrogen and phosphorus concentrations in the treated effluents range from 3 to 20 mg L\textsuperscript{-1} and 3 to 5 mg L\textsuperscript{-1}, respectively.\textsuperscript{21} According to the Queensland Water Quality Guidelines for total nitrogen and phosphorous in freshwaters, they should be less than 0.5 and 0.05 mg L\textsuperscript{-1}, respectively.\textsuperscript{22}
There are many pollutants commonly associated with stormwater runoff, wastewater and treated effluent with nutrients as one of the pollutants of concern. Nutrients in aquatic systems come from natural processes and human activities. Natural sources include weathering processes of rock, fixation of atmospheric nitrogen by bacteria and leguminous plants, decomposition of organic material, and soil leaching. Human activity sources consist of fertilisers, human waste, and industrial discharge and sewage.

1.2 Nutrients

1.2.1 Nitrogen and Phosphorus

Significant and widespread nitrogen and phosphorus pollutions have been reported in many water bodies globally\textsuperscript{23-28} (Table 1-1) and excessive nutrient loadings to estuaries, rivers and lakes are one of the major concerns for water quality management.\textsuperscript{29} Natural waters contain significant levels of dissolved inorganic nitrogen [DIN, including ammonium (NH\textsubscript{4}-N), nitrate (NO\textsubscript{3}-N), and nitrite (NO\textsubscript{2}-N)] as well as dissolved organic nitrogen (DON).\textsuperscript{30}

Phosphorus in aquatic systems occurs in three forms: dissolved inorganic phosphorus (DIP), particulate organic phosphorus (POP), and dissolved organic phosphorus (DOP). The orthophosphate ion (PO\textsubscript{4}-P) is the most significant form of inorganic phosphorus, and the only form of soluble inorganic phosphorus directly utilised by aquatic biota. Orthophosphate makes up a significant portion of the commonly measured dissolved reactive phosphorus (DRP) fraction (also referred to as “filterable reactive phosphorus”).\textsuperscript{31} Most studies of nutrients in water focus on DIN and PO\textsubscript{4}-P, which are the most readily available N and P sources for aquatic plants and bacteria.\textsuperscript{32, 33}
<table>
<thead>
<tr>
<th>Location</th>
<th>Year</th>
<th>Description</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>The Rio Grande of central New Mexico, USA</td>
<td>1989-1997</td>
<td>Ammonia concentrations exceeded legislated values 97%, 74%, 78%, and 11% of the time in 1989, 1991, 1992, and 1997, respectively in USA. High ammonium concentrations occurred in rivers was because the sewage treatment capabilities were poorly developed or absent.</td>
<td>24</td>
</tr>
<tr>
<td>Europe</td>
<td>1985-2000</td>
<td>Surface water contamination with phosphorus was a major problem in lowland Europe due to the diffuse pollutions from land. Although the targeted point sources have been reduced, high total phosphorus concentrations (0.1 - 0.3 mg L$^{-1}$) were found in 80% of the sandy regions in the Netherlands.</td>
<td>27</td>
</tr>
<tr>
<td>Texas groundwater in USA</td>
<td>2000</td>
<td>Over 50% of the observations in west-central and north-central Texas were higher than the maximum contaminant level (MCL) of 50 mg L$^{-1}$ of nitrate. The highest nitrate concentration was discovered in the Seymour Aquifer, which was 59.9 mg L$^{-1}$.</td>
<td>23</td>
</tr>
<tr>
<td>Australia</td>
<td>2003</td>
<td>High groundwater nitrate concentrations were identified in all states and territories, across various land uses. Nitrate concentrations in two inland areas of Northeast Australia: the Eastern Darling Downs and the Callide Valley were above 50 mg L$^{-1}$. High nitrate concentration was also found in irrigated areas of Northern Victoria, horticultural areas overlaying the coastal aquifers near Perth and under irrigated and dryland pastures and vineyards of the southeast region of South Australia.</td>
<td>26</td>
</tr>
<tr>
<td>Haihe River Basin, China</td>
<td>2010</td>
<td>Ammonium concentrations in 21 monitoring sections were from 0.435 to 58.77 mg L$^{-1}$. The levels of ammonium in Dagu Drainage River, Beitang Drainage River and Ziya were 24.7, 19.5 and 17.4 times higher than Chinese national water quality V standard (2.0 mg L$^{-1}$), respectively.</td>
<td>25</td>
</tr>
<tr>
<td>Lake Taihu, China</td>
<td>2010</td>
<td>The average total phosphorus concentration in lake Taihu remained at 0.086 mg L$^{-1}$. The highest concentration was found in northern part of the lake which was up to 0.145 mg L$^{-1}$ due to the widespread use of fertiliser.</td>
<td>28</td>
</tr>
</tbody>
</table>
Nutrient concentrations in natural waters are usually low, for example, ammonium concentrations are typically below 0.2 mg L⁻¹, nitrate less than 1 mg L⁻¹, nitrite below 0.1 mg L⁻¹, and phosphorus down to 0.01 mg L⁻¹ or even lower. However, inorganic nutrients can reach high levels as a result of agricultural runoff, sewage discharge and human or animal wastes. With the widespread use of fertilisers and release of treated or untreated sewage, nitrogen and phosphorus concentrations in surface and ground waters have increased dramatically worldwide. The resultant elevated nutrient loads to rivers, lakes, bays and estuaries have led to eutrophication and the associated problems of phytoplankton and algal blooms, and subsequent water column hypoxia or anoxia when these blooms collapse and decompose. Therefore, the ecological health of freshwater and coastal areas has become a global concern. Liu et al. modelled the past and future inorganic N and P in the world’s major rivers (Figures 1-1 and 1-2). The results indicate that the number of rivers polluted by DIN and DIP increased by 33% and 25%, respectively, from 1970 to 2000 and predicted that DIN and DIP will increase by a further 38% and 77%, respectively, by 2050. Therefore, this study will focus on developing techniques to monitor DIN (NH₄-N, NO₃-N and NO₂-N) and DIP (PO₄-P) in freshwaters.
**Figure 1-1.** Water pollution levels of major rivers for DIN in 1970, 2000, 2030 GO (from Liu et al. 2012). The numbers of rivers polluted by DIN have increased from 1970 to 2000, and this trend will continue to increase until 2030 and 2050.
The numbers of rivers polluted by DIP have increased from 1970 to 2000, and this trend will continue to increase until 2030 and 2050.

1.2.2 Impacts of nitrogen and phosphorus pollution on the environment and humans

Ammonia is toxic to fish and other aquatic fauna, although the degree of toxicity differs significantly among species, and with life stage and salinity. Ammonia can damage the gill epithelium causing asphyxiation, stimulate glycolysis and suppress the Krebs cycle leading to progressive acidosis, which reduces the oxygen-carrying capacity of the blood, disrupts blood vessels, impacts osmoregulatory activity and affects liver and kidney functions. High levels of nitrate can potentially cause the death of fish, invertebrates and amphibians due to the conversion of oxygen-carrying pigments to forms that are incapable of carrying oxygen. Nitrate levels above 30 mg L$^{-1}$ can inhibit growth, impair the function of the immune system and lead to stress in some aquatic species. Phosphorus can also be toxic, but this toxicity
rarely occurs in nature, due to its generally low concentration. In addition to their direct toxic effects, high concentrations of nitrogen and phosphorus stimulate phytoplankton and algal growth in water bodies which can ultimately lead to water column hypoxia and anoxia, and consequent mortality amongst fauna when their blooms collapse and decompose. Repetitive algal blooms can cause highly undesirable changes in ecosystem structure and function.38

In humans, high concentrations of nitrate/nitrite (NOx) in drinking water can pose a serious threat, as they can cause severe health problems.23,38-41 The major biological effect in humans is involvement of NOx in the oxidation of haemoglobin to methaemoglobin (MetHb), which is unable to transport oxygen to the tissues. Symptoms include cyanosis, headaches, dizziness, fatigue, seizures, coma and death depending on the MetHb concentration.42, 43 Infants younger than six months are particularly sensitive to NOx-induced methemoglobinemia giving this condition its name of “blue-baby syndrome” due to the cyanosis that occurs.22, 38, 43, 44 Additionally, it has been postulated that excess NOx in drinking water is responsible for increased rates of various cancers in humans, such as esophageal, gastric and nasopharyngeal cancer.38 Phosphate is an important substance in the human body as a component of DNA and RNA, and energy metabolism. High concentrations of phosphorus, however, can cause kidney damage and osteoporosis.45 Several studies suggest that high intakes of phosphorus are also associated with an increased risk of cardiovascular disease.46, 47

1.2.3 Current water quality guidelines for nitrogen and phosphorus

Ammonium/ammonia is often the dominant form of dissolved inorganic nitrogen in natural waters. Unionised ammonia (NH₃) is much more toxic than the ammonium ion (NH₄⁺),48-50 because it is a neutral molecule that is able to diffuse freely across the epithelial membranes of aquatic organisms. Additionally, the ammonium ion and unionised ammonia are easily
interchangeable, with the ratio of ammonia to ammonium largely depending on pH, temperature and salinity.\textsuperscript{51} However, at the circa neutral pH of most natural waters, ammonium is present at much greater concentrations than ammonia.

Many countries use unionised ammonia as a parameter for water quality while some countries use ammonium and total ammonia/ammonium. Ammonia is not of immediate health relevance in drinking water, therefore, the World Health Organization (WHO) does not propose any health-based guideline value.\textsuperscript{3} Not many counties have guidelines for ammonia and ammonium in drinking water due to the low concentrations in raw and treated water.\textsuperscript{52, 53} In Australia, there is no health-based guideline value for ammonia but there is an aesthetic guideline for ammonia in drinking water of 0.41 mg L\textsuperscript{-1}.\textsuperscript{54} The EU has established a guideline for ammonium in drinking water which is 0.23 mg L\textsuperscript{-1}.\textsuperscript{55} Many countries have set up the guidelines for unionised ammonia and total ammonia in natural water bodies.\textsuperscript{22, 48, 56, 57} Canada has established the lowest criterion of 0.019 mg L\textsuperscript{-1} unionised ammonia,\textsuperscript{48} followed by the EU with 0.025 mg L\textsuperscript{-1} and Australia with 0.02 mg L\textsuperscript{-1}.\textsuperscript{22, 57} For total ammonia, the EU has the lowest criterion of 1 mg L\textsuperscript{-1},\textsuperscript{57} followed by the USA with 1.9 mg L\textsuperscript{-1} (at 20 °C and pH of 7.0)\textsuperscript{56} and Canada with 3.97 mg L\textsuperscript{-1} (at 20°C and pH of 7.0).\textsuperscript{48} Additionally, the EU also has guidelines for guidance for unionised ammonia and total ammonium in freshwater, which are 0.005 mg L\textsuperscript{-1} and 0.2 mg L\textsuperscript{-1}, respectively.\textsuperscript{57}
Table 1-2. Current water quality guidelines for unionised ammonia, ammonium and total ammonia.

<table>
<thead>
<tr>
<th>Source</th>
<th>Unionised ammonia (N)</th>
<th>Ammonium (N)</th>
<th>Total ammonia (N)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Drinking water guidelines</strong></td>
<td></td>
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<tr>
<td>World Health Organization (WHO) Guideline for Drinking Water Quality</td>
<td>-</td>
<td>-</td>
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<td>3</td>
</tr>
<tr>
<td>Australian Drinking Water Guidelines</td>
<td>0.41 mg L⁻¹ (aesthetic)</td>
<td>-</td>
<td>-</td>
<td>54</td>
</tr>
<tr>
<td>Guidelines for Canadian Drinking Water Quality</td>
<td>-</td>
<td>-</td>
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<td>52</td>
</tr>
<tr>
<td>USEPA National Primary Drinking Water Standards (USA)</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>53</td>
</tr>
<tr>
<td>European Communities Drinking Water Regulations</td>
<td>-</td>
<td>0.23 mg L⁻¹</td>
<td>-</td>
<td>55</td>
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<tr>
<td><strong>Freshwater guidelines</strong></td>
<td></td>
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<tr>
<td>Australian and New Zealand Guidelines for Fresh and Marine Water Quality</td>
<td>0.02 mg L⁻¹ (cold water)</td>
<td>0.02 mg L⁻¹ (south-east Australia)</td>
<td>22</td>
<td></td>
</tr>
<tr>
<td>Canadian Water Quality Guidelines for the protection of Aquatic Life</td>
<td>0.03 mg L⁻¹ (warm water)</td>
<td>3.97 mg L⁻¹ (20°C and pH 7.0)</td>
<td>48</td>
<td></td>
</tr>
<tr>
<td>USEPA National Recommended Water Quality Criteria (USA)</td>
<td>0.019 mg L⁻¹</td>
<td></td>
<td></td>
<td>56</td>
</tr>
<tr>
<td>Directive 2006/44/EC of the European Parliament and of the Council on the Quality of Fresh Waters Needing Protection or Improvement in Order to Support Fish Life</td>
<td>0.05 mg L⁻¹ (guidance)</td>
<td>17 mg L⁻¹ (acute)</td>
<td>57</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.025 mg L⁻¹ (imperative value)</td>
<td>1.9 mg L⁻¹ (chronic 20°C and pH 7.0)</td>
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<td></td>
<td>0.2 mg L⁻¹ (guidance)</td>
<td>0.2 mg L⁻¹ (guidance)</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>1 mg L⁻¹ (imperative value)</td>
<td>1 mg L⁻¹ (imperative value)</td>
<td></td>
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</tbody>
</table>
The WHO has established drinking water guidelines for nitrate ($11.3 \text{ mg L}^{-1}$) and nitrite [0.91 mg L$^{-1}$ (short-term) and 0.061 mg L$^{-1}$ (long-term)].$^3$ Most countries have established drinking water guidelines for nitrate, which are very similar (approximately $10 \text{ mg L}^{-1}$) across different geographic areas.$^{52-55}$ Most countries, except the EU, also have the similar guidelines for nitrite in drinking water, which are approximately $1 \text{ mg L}^{-1}$. The EU has the lowest guideline for nitrite of $0.15 \text{ mg L}^{-1}$. In freshwaters, most countries, except the USA,$^{49}$ have the similar guidelines for nitrate (between $11.3$ and $13 \text{ mg L}^{-1}$) and nitrite (between $0.03$ and $0.06 \text{ mg L}^{-1}$).$^{22, 48, 58}$ In addition, there is a short-term freshwater guideline for nitrate in Canada, which is up to $550 \text{ mg L}^{-1}$.$^{48}$

No countries have established guidelines for dissolved reactive phosphorus or total phosphorus in drinking water because of its uniformly low concentration.$^{3, 52-55}$ Australia and Canada have set guidelines for total phosphorus in freshwaters,$^{22, 48}$ while the EU has established guidelines based on dissolved reactive phosphorus.$^{58}$ Additionally, south-east Australia has regional guideline trigger values for dissolved reactive phosphorus and total phosphorus in freshwater of $0.02 \text{ mg L}^{-1}$ and $0.05 \text{ mg L}^{-1}$, respectively.$^{22}$
Table 1-3. Current water quality guidelines for nitrate and nitrite.

<table>
<thead>
<tr>
<th>Source</th>
<th>Nitrate (N)</th>
<th>Nitrite (N)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Drinking Water Guidelines</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>World Health Organization (WHO) Guideline for Drinking Water Quality</td>
<td>11.3 mg L$^{-1}$</td>
<td>0.91 mg L$^{-1}$ (short-term)</td>
<td>3</td>
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<tr>
<td></td>
<td></td>
<td>0.061 mg L$^{-1}$ (long-term)</td>
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<tr>
<td>Australian Drinking Water Guidelines</td>
<td>11.3 mg L$^{-1}$</td>
<td>0.91 mg L$^{-1}$</td>
<td>54</td>
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<tr>
<td>Guidelines for Canadian Drinking Water Quality</td>
<td>10 mg L$^{-1}$</td>
<td>1 mg L$^{-1}$</td>
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<tr>
<td>USEPA National Primary Drinking Water Standards (USA)</td>
<td>10 mg L$^{-1}$</td>
<td>1 mg L$^{-1}$</td>
<td>53</td>
</tr>
<tr>
<td>European Communities Drinking Water Regulations</td>
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<td>0.15 mg L$^{-1}$</td>
<td>55</td>
</tr>
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<td><strong>Freshwater guidelines</strong></td>
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<tr>
<td>Australian and New Zealand Guidelines for Fresh and Marine Water Quality</td>
<td>11.3 mg L$^{-1}$</td>
<td>0.03 mg L$^{-1}$</td>
<td>22</td>
</tr>
<tr>
<td>Canadian Water Quality Guidelines for the protection of Aquatic Life</td>
<td>550 mg L$^{-1}$ (short-term)</td>
<td>0.06 mg L$^{-1}$</td>
<td>48</td>
</tr>
<tr>
<td></td>
<td>13 mg L$^{-1}$ (long-term)</td>
<td></td>
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</tr>
<tr>
<td>USEPA National Recommended Water Quality Criteria (USA)</td>
<td>-</td>
<td>-</td>
<td>49</td>
</tr>
<tr>
<td>EU-wide Groundwater Quality Standards</td>
<td>11.3 mg L$^{-1}$</td>
<td>0.03 mg L$^{-1}$</td>
<td>58</td>
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</tbody>
</table>
Table 1-4. Current water quality guidelines for phosphorus.

<table>
<thead>
<tr>
<th>Source</th>
<th>Dissolved reactive phosphorus (P)</th>
<th>Total phosphorus (P)</th>
<th>References</th>
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<tbody>
<tr>
<td><strong>Drinking Water Guidelines</strong></td>
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<tr>
<td>World Health Organization (WHO) Guideline for Drinking Water Quality</td>
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<td>3</td>
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<td>Australian Drinking Water Guidelines</td>
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<tr>
<td>Guidelines for Canadian Drinking Water Quality</td>
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<td>USEPA National Primary Drinking Water Standards (USA)</td>
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<td><strong>Freshwater guidelines</strong></td>
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</tr>
<tr>
<td>Australian and New Zealand Guidelines for Fresh and Marine Water Quality</td>
<td>0.02 mg L(^{-1}) (south-east Australia)</td>
<td>0.03 mg L(^{-1}) (Australia)</td>
<td>22</td>
</tr>
<tr>
<td>Canadian Water Quality Guidelines for the protection of Aquatic Life</td>
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<td>48</td>
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<tr>
<td>USEPA National Recommended Water Quality Criteria (USA)</td>
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<td>56</td>
</tr>
<tr>
<td>EU-wide Groundwater Quality Standards</td>
<td>0.2 mg L(^{-1}) (eutrophic waters)</td>
<td>-</td>
<td>58</td>
</tr>
<tr>
<td></td>
<td>0.1 mg L(^{-1}) (mezotrophic waters)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.04 mg L(^{-1}) (oligotrophic waters)</td>
<td></td>
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</tr>
</tbody>
</table>

\(^a\) Canadian Guidance Framework provides trigger ranges for total phosphorus (µg L\(^{-1}\))
1.3 Monitoring of nutrient concentrations

To assess the quality of water sources and the likelihood of eutrophication in freshwaters, nutrient concentrations have to be monitored. Nutrient concentrations can change dramatically from place to place due to differences in weather and especially run-off,\(^{59}\) land use,\(^{26}\) geology, water flow, the presence/absence of point sources (e.g. wastewater treatment plants), the relative dominance of N-cycling processes (e.g. nitrification and denitrification), and seasonal and diurnal shifts between net autotrophy (production) and heterotrophy.\(^{60-62}\) Additionally, the accuracy of concentration measurements and load estimates depends on the sampling frequency and representativeness of sampling.\(^{63}\) The monitoring techniques for nutrients include grab sampling, on-site sampling and *in situ* sampling. The advantages and disadvantages of these sampling strategies are presented and compared below.

### 1.3.1 Grab sampling technique

The nutrient concentrations determined by monitoring agencies for compliance purpose in many countries in the world are often based on grab samples, which are infrequent sampling data\(^ {64-67}\) as samples are collected from the sampling sites at fixed time intervals, e.g. weekly or monthly.\(^ {64}\) This approach, however, can only at best provide a snapshot of the concentrations of nutrients at the moment of sampling, and sampling frequencies are generally insufficient to capture the dynamic behaviour of water quality. As nutrient concentrations change continuously over time in most water systems, the results from grab sampling are unlikely to be representative of actual conditions.\(^ {68}\) Increased sampling frequencies can reduce this problem, but this increases labour and time costs.
1.3.2 On-site measurement

On-site monitoring is a more rapid approach where analytes are measured automatically or manually close to the sample site such as on the shore of a lake, river bank, or on board a ship. This approach involves field based analytical techniques that are adapted from those usually carried out within the laboratory. On-site monitoring results in more rapid analysis of the samples, and thus minimises changes in the samples such as shifts in analyte speciation.

On-site monitoring is close to real-time analysis, however, it does have some limitations. Firstly, results obtained in the field may be less accurate than laboratory analyses because the adaptation of measurement methods to the field results in lower reproducibility and poorer detection limits compared to the equivalent laboratory technique. Moreover, the quality assurance measures routinely implemented in laboratory analytical procedures can be difficult to incorporate into on-site monitoring programs, which may affect the integrity of the results. Finally, the equipment for on-site monitoring is often expensive to purchase and maintain, and field installation requires a sheltered environment with an electrical power supply. Therefore, although on-site monitoring can provide considerable advantages over conventional grab sampling techniques, particularly due to the higher sampling frequency, these limitations have restricted its use for monitoring applications.

1.3.3 In situ measurement

In situ monitoring of nutrients has been of great importance in many environmental applications, particularly those involving biological treatment processes and eutrophication monitoring. In situ monitoring includes direct measurement of contaminants in the environment or direct accumulation of contaminants in stable forms from the environment followed by laboratory analysis. For instance, the use of sensing devices directly immersed in the water bodies allows data to be collected automatically, recorded for later retrieval or
transmitted telemetrically to a distant laboratory for interpretation. Glibert et al. employed *in situ* nutrient monitors to capture nutrient variability and its relationship with algal blooms in Chesapeake Bay.\(^7\) The data were collected in time-series (every 2 - 3 h) to document short-term variability. However, the challenges of application of *in situ* nutrient sensors in natural waters are numerous. Firstly, low-level detection limits can be a major concern for *in situ* nutrient sensors, which cannot measure nutrient concentrations lower than 0.1 mg L\(^{-1}\).\(^7\) Additionally, they require a large dynamic range over periods of several-day, and are subject to interference from suspended material and dissolved humics.\(^7\) A further issue for *in situ* sensors is the growth of biofilms on the sensor, which results in the measurement of the biologically modified concentrations in the biofilms rather than bulk water column values.

*In situ* sampling techniques can be divided into three broad categories: continuous, discrete, and time-integrated methods.\(^7\) The type of data obtained by each of these method categories is summarised in Figure 1-3. Continuous *in situ* sampling represents the ideal method for all applications due to its ability to obtain a real time representation of variations in analyte concentrations. These techniques are often expensive and the equipment used can be sensitive, fragile or subject to artifacts e.g. due to growth of biofilms on sensors.\(^6\) Discrete sampling techniques, much like conventional grab samples, provide a good representation of concentrations but only at the time of measurement and they cannot provide information about the concentrations between the measurement points.\(^7\) In addition, discrete *in situ* measurements require more frequent sampling times compared with the time-integrated approach\(^7\) (e.g. in Figure 1-3, nine points would be required to measure the deployment period compared to only four deployments for the time-integrated approach). Time-integrated *in situ* techniques provide a number of advantages compared with continuous and discrete *in situ* sampling, which are discussed further below.
1.3.4 In situ passive sampling techniques

Time-integrated in situ (also named passive sampling) techniques determine the average analyte concentration over a known deployment time. Since the time-integrated measurement responds to the changes in concentration over each deployment, it integrates temporal environmental variability, thereby providing a more representative measure of variations in contaminant concentrations compared with discrete sampling techniques. Moreover, because the analyte species are continuously accumulated within the passive sampling device, their concentrations in the extract will be higher than that obtained using grab sampling, reducing the uncertainty of analytical determination and greatly improving detection limits. Additionally, the time-integrated measurement requires less labour as the
passive sampling device can be deployed in the field for periods ranging from hours to days rather than the collection of samples every hour or day using grab sampling techniques.

Rozemeijer et al. developed a SorbiCell-sampler — a passive sampling technique for measuring nitrate and phosphate in surface waters and drain effluents. The SorbiCell sampler (SC-sampler) can measure average concentrations over long periods of time (days-months) for various substances. The method is based on the advective flow of water through the sampler, rather than equilibrium or kinetic diffusion. To induce the water flow through the SC-sampler, they are usually placed on reservoirs with atmospheric pressure inside. However, this method may not accurately measure the low concentrations of nitrate in water due to its high relative standard deviation.

Several diffusive gradients in thin films (DGT) techniques have been developed for determination of phosphate (PO$_4$-P) in water, including ferrihydrite-DGT, Metsorb-DGT and Zr-oxide-DGT. The DGT technique is a passive sampling method which is further described in the following section. Panther et al. developed a DGT technique for measuring phosphate in fresh and marine waters using titanium dioxide (Metsorb) as a binding agent. Metsorb-DGT is independent of pH from 3.5 to 8.5 and has a high binding capacity for phosphate. The Metsorb-DGT method has the potential to be used to monitor phosphate in natural waters, but still needs to be systematically evaluated in the field, which is one of the aims of this study.

In terms of the representativeness of different sampling techniques, passive sampling is better than low frequency grab sampling and on-site sampling. Although intensive grab sampling can still provide representative results of analytes in water systems, this will increase labour
and time costs. Passive sampling techniques, however, have relatively high levels of uncertainty in the sample collection stage compared with the other two techniques. This is because the fraction of analyte measured by passive samplers can differ depending on the speciation of the analytes, especially for metals. To reduce the level of uncertainty, passive samplers need to be better characterised to increase their affinity for the target analyte species. Although this may be a challenge, it can significantly reduce the uncertainty associated with the sample collection stage.

1.4 The diffusive gradients in thin films (DGT) technique

The DGT technique is a well-establish time-integrated, in situ, passive sampling technique, that has commonly been utilised to measure concentrations of various contaminants in water, including trace metals and phosphorus in rivers, lakes, estuaries and coastal waters. However, currently there are no DGT techniques that can measure the concentrations of inorganic nitrogen species in water. The development of DGT techniques capable of measuring ammonium and nitrate could provide a reliable and robust method for the routine monitoring of these nutrients in dynamic freshwater systems.

1.4.1 DGT theory

The DGT technique relies on the diffusion of the analyte(s) though a hydrogel of known thickness to a second hydrogel layer containing an analyte specific binding agent. The standard DGT probe is made from plastic and consists of a circular cap with a 2.00-cm-diameter open window, a protective covering filter membrane, a diffusive gel of known thickness (Δg), an analyte specific binding gel and a piston (Figure 1-4). When an appropriate diffusive gel thickness is selected, the rate of transport of the analyte to the binding gel layer is solely restricted to molecular diffusion. The diffusive gel layer is commonly made
from polyacrylamide that allows diffusion of the analyte and establishment of a steady-state concentration gradient between the bathing solution and the analyte specific binding agent in the binding gel layer. The successive binding gel layer incorporates an analyte selective binding agent, which preconcentrates and immobilises the analyte. Analytes in the bulk solution must initially diffuse through a zone of thickness (δ) known as the diffusive boundary layer (DBL) at the device surface before entering the diffusive gel layer. Mass transport in this DBL is also restricted to molecular diffusion. The thickness of the DBL is affected by the flow rate in the bulk solution as the dominant factor, and will extend ∆g from the face of the DGT sampler out into the bulk solution. However, when the flow velocity is greater than 0.02 m s⁻¹, δ is considered to be negligible allowing diffusive transport to be dominated by ∆g. Thus as long as the diffusive gel layer is of sufficient thickness and the flow rate is adequate, the influence of the solution hydrodynamics on the diffusion of the analyte into the DGT probe is insignificant. Figure 1-5 displays the equilibrium linear diffusion gradient that is established within a DGT device in water. The diffusive layer is shown as a single layer which consists of the DBL, filter membrane and diffusive gel layer.
Figure 1-4. Schematic illustration of the basic components of a DGT device.

Figure 1-5. Graphical representation of the diffusion gradient established within a DGT probe in contact with a solution (modified from Davison and Zhang 1994).
The analyte concentration in solution is calculated using Fick’s first law of diffusion and the measured mass of the analyte accumulated on the binding agent after a known deployment.\textsuperscript{79, 87, 88} The flux ($F$) of solute from the bulk solution to the binding phase can be expressed as:

$$ F = \frac{D(C - C')}{\Delta g} $$ \hspace{1cm} (1)

Where $D$ is the diffusion coefficient of the analyte in the hydrogel, $C$ is the concentration of the analyte in the bulk solution, $\Delta g$ is the diffusional path length (thickness of the diffusive gel layer and filter membrane) and $C'$ is the concentration of analyte in the binding gel layer. If the analyte is in rapid equilibrium with a binding agent with a strong binding constant, it is assumed that $C'$ is effectively zero, provided that the binding agent is not saturated, as the binding layers will no longer quantitatively accumulate the analyte when saturated. Therefore equation (1) can be simplified to:

$$ C = \frac{F \Delta g}{D} $$ \hspace{1cm} (2)

$F = \frac{M}{At}$, where $F$ is the flux of the solute, $M$ is the mass of the analyte accumulated by the binding gel, $A$ (3.14 cm$^2$) is the area of gel exposed to the solution, and $t$ is the exposure time. The analyte mass accumulated by the binding agent ($M$) can be quantified by elution of the analyte from the binding gel layer and analysis of the concentration of the analyte in a known volume of eluent.\textsuperscript{79, 87, 88} Hence, equation (1) can then be solved for $C$ to provide a time-integrated measure of the solution concentration of the analyte.\textsuperscript{87}

$$ C = \frac{M \Delta g}{DtA} $$ \hspace{1cm} (3)
In waters with insufficient flow, the DBL cannot be ignored and needs to be calculated. To achieve this, DGT samplers with various diffusive layer thicknesses are deployed so that the thickness ($\delta$) of the DBL can be determined.\textsuperscript{89, 90}

\[
\frac{1}{M} = \frac{\Delta g}{DC_{DGT}At} + \frac{\delta}{DC_{DGT}At} \quad (4)
\]

A plot of $1/M$ versus $\Delta g$ yields a straight line with a slope ($m$) of $1/(DC_{DGT}At)$ and intercept ($b$) of $\delta/(DC_{DGT}At)$. Therefore, $\delta$ (Eq. 5) and $C_{DGT}$ (Eq. 6) can be calculated.

\[
\delta = \frac{b}{m} \quad (5)
\]

\[
C_{DGT} = \left(\frac{1}{mDAt}\right) \quad (6)
\]

When the thickness of the DBL is included in the DGT calculations, a value of 3.8 cm\textsuperscript{2} is used for the area exposed to solution $A$, as the effective sampling window has a larger diameter (2.20 cm) than the geometric diameter of the exposure window (2.00 cm) due to the possibility of lateral diffusion within the gels (See Warnken \textit{et al.}).\textsuperscript{89}

1.4.2 DGT binding agents

A number of different binding agents, including Chelex-100, ferrihydrite, and Metsorb [titanium dioxide (TiO\textsubscript{2})], have been applied for DGT measurements.

Chelex-100 is the most commonly used binding agent and is able to bind divalent and trivalent metal ions in the presence of high concentrations of alkali metals such as in
seawater.\textsuperscript{91} Chelex-100 is a chelating resin with iminodiacetic acid functional groups and its efficiency in binding metals depends on solution pH.\textsuperscript{83, 91, 92} Additionally, natural waters with low cation concentrations could affect the quantitative measurements of Chelex-DGT for trace metals.\textsuperscript{86, 93, 94} A number of studies have also investigated the development and optimisation of Chelex-100 resin for the adsorption of lanthanide species.\textsuperscript{91}

Ferrihydrite was identified by Zhang \textit{et al.} for the measurement of phosphate in natural waters.\textsuperscript{79} Ferrihydrite has also been used by Panther \textit{et al.} to measure inorganic arsenic in water.\textsuperscript{95} However, there are issues associated with the ferrihydrite binding phase. Firstly, it requires careful laboratory synthesis, which, if not controlled adequately, can produce a mixture of ferrihydrite, goethite and hematite with a progressive conversion of ferrihydrite to goethite, which results in inconsistent measurements and deteriorating uptake efficiency. Secondly, the procedure for synthesising ferrihydrite is time-consuming, tedious, and susceptible to error.

A new binding agent, an agglomerated nano-crystalline titanium dioxide based adsorbent (Metsorb) was developed by Bennett \textit{et al.} and validated as a DGT adsorbent for the measurement of total inorganic arsenic and selenium in natural waters.\textsuperscript{81} The Metsorb-DGT has also been evaluated and utilised to monitor other oxyanions, including phosphate,\textsuperscript{32} aluminium,\textsuperscript{92} and uranium\textsuperscript{96} in freshwaters and seawater. This titanium dioxide-based adsorbent has been proved to be a good alternative to ferrihydrite for accumulating phosphate due to its higher binding capacity and better performance in natural waters.\textsuperscript{97}
1.4.3 Advantages of the DGT techniques

The DGT technique has a number of advantages as a monitoring tool. Firstly, the DGT technique is a low energy requirement technique which does not need any power source, and uses less labour. Secondly, the DGT technique is a passive sampling technique that facilitates measurement of time-weighted average concentrations of a range of elements. The DGT technique uses an analyte-specific binding gel layer to accumulate analytes in situ and allows quantification using sensitive laboratory instrumentation. The time-integrated measurement can provide a more representative measurement which is superior to discrete measurements.

DGT preconcentrates the analyte of interest to much higher levels compared to the bulk solution concentration, because the analyte is accumulated within the binding gel layer. Therefore, DGT can be utilised to determine trace and ultra-trace concentrations of contaminants, which may not have been detectable via conventional methods. Preconcentration of the analyte within the binding gel layer can also greatly reduce matrix interferences (e.g. Ca$^{2+}$ and Cl$^-$ in seawater), as after elution of the analyte from the binding phase, the eluent can be diluted to a concentration that does not interfere with analysis.

Finally, the DGT technique can provide a method for measuring the speciation of particular analytes depending on their liability. For instance, Chelex-100 only binds free metal ions, thus metal complexes can only be determined if they can dissociate within the measurement time, and those species which cannot dissociate within the measurement time are not measured. Additionally, the diffusive gel pore size and structure can be easily manipulated so only certain species of analytes are able to pass through. For instance, DGT probes with diffusive gels with large pore size allow both small inorganic species and larger organically
complexed species to diffuse to the binding layer, whilst diffusive gels with restricted pore size limit diffusion to the smaller inorganic species.\textsuperscript{85}

1.4.4 Limitations of the DGT techniques

Although the DGT technique has a number of advantages, it also has some limitations. These limitations are related to the environmental conditions in which the DGT probes are deployed. The first potential limitation recognised during the development of the DGT technique by Zhang and Davison was related to biofouling of the exposed surface of the DGT probes.\textsuperscript{88} Biofilms may develop on DGT probes when algae and bacteria grow. Suspended particulates, which contain micro-organisms, can also adhere to the biofilms or exposed surfaces and promote biofilm growth on the surface of the probe.\textsuperscript{102} The presence of a biofilm on a DGT probe interferes with DGT measurement in two ways. Firstly, the biofilm increases the diffusional path length for the analyte, leading to an underestimation of the solution concentration. Secondly, biological processes within the biofilm can cause changes in the concentration and speciation of the analytes.\textsuperscript{102} Pichette \textit{et al.} investigated a number of agents that can prevent the formation of biofilms on DGT probes and found that treatment of the filter membrane with metals such as silver and copper could prevent the development of biofilms. This method, however, can only be used when sampling non-metals such as nutrients, where the presence of the metals themselves will not interfere measurements.\textsuperscript{102}

The problem of biofouling is most prevalent in water with high organic, nutrient and/or suspended particulate levels. Therefore, using DGT probes under these conditions is somewhat limited, as the maximum deployment time is limited by the rapid growth of biofilms. Pichette \textit{et al.} also investigated the concentration changes of phosphorus measured
by DGT devices over different deployment times at high nutrient levels. It was reported that the phosphorus concentration in the field was underestimated by the DGT technique over deployments longer than four days due to the growth of biofilms.\textsuperscript{102} Therefore, the deployment times in this study were restricted to less than four days.

A further limitation of DGT methods is associated with the diffusive boundary layer.\textsuperscript{88} A DBL, of thickness \( \delta \), forms at the surface of the deployed DGT probe separating the exposed probe surface from the bulk solution by a layer of unmixed water.\textsuperscript{87} Within this layer, the transport of solutes is controlled solely by molecular diffusion, therefore, the presence of a DBL essentially increases the distance that solutes must diffuse to reach the binding layer. This subsequently influences the estimation of solution concentration, as the diffusive path length is greater than \( \Delta g \).\textsuperscript{87} The thickness of the DBL is affected by the flow rate of the bulk solution and becomes increasingly important as this rate decreases.\textsuperscript{101} Gimpel \textit{et al.} found that when the flow velocity of the bulk solution was greater than 0.02 m s\textsuperscript{-1}, the effect of the DBL became negligible for diffusive gels thicknesses of \( \geq 0.7 \) mm (Figure 1-6).\textsuperscript{101}
The ratio of DGT measured Cd to Cd measured in the bulk solution for two designs of holders, standard (●) and fluted (〇), at various flow velocities (Gimpel et al. 2001).101

Normally, the flow rate of rivers and streams is greater than 0.02 m s⁻¹, and there is sufficient natural convection in most lakes and oceans to achieve this velocity, which means that the DGT technique can be used in most systems. However, the application of DGT in stationary water bodies like poorly mixed lakes and ponds may require the deployment of DGT probes with three common diffusive gel layer thickness (∆g = 0.05, 0.09 and 0.13 cm) to allow for the calculation of the DBL thickness and subsequent correction for its influence on the estimate of the solute concentration.

Finally, low ionic strength may affect the DGT performance.93 Some studies have suggested that when DGT devices are deployed in water of very low ionic strength (< 10⁻³ mol L⁻¹), the effective diffusion coefficients of some analyte metals increased.86 Although low ionic
strength may affect the behaviour of DGT, Peters et al. pointed out that it would not affect most measurements made by DGT, as above an ionic strength of 0.1 mmol L$^{-1}$, DGT worked predictably for Ca, Cd, Co, Cr (III), Cu, Mg, Ni, Pb and Zn and conformed to the theory of Eq. (5).$^{93,103}$ Therefore, in the vast majority of natural waters, DGT can be expected to work reliably as both a device for measuring concentrations of labile analytes and a speciation tool.$^{93,103}$

1.5 Objectives

This research aims to develop new DGT techniques for the measurement of dissolved inorganic nitrogen (nitrate and ammonium) in freshwater and stormwater. It also aimed to utilise these new DGTs and the existing Metsorb-DGT for phosphate to determine representative dissolved inorganic nitrogen and phosphorus concentrations in freshwaters. More specially, the primary objectives of the study are to:

1. Develop, characterise and evaluate a diffusive gradients in thin films (DGT) technique to measure nitrate in freshwaters.

2. Develop, characterise and evaluate a diffusive gradients in thin films (DGT) technique to measure ammonium in freshwaters.

3. Systematically validate the new DGT methods and the existing Metsorb-DGT technique to monitor concentrations and concentration changes of dissolved inorganic nutrients in various aquatic systems. The DGT deployments were accompanied by the frequent collection of grab water samples for nutrients, to allow direct comparison of
the time-weighted average DGT concentrations to those determined in the grab samples.

1.6 Content and structure of the thesis

This thesis consists of six chapters: the Introduction (Chapter 1), four results chapters (Chapters 2 - 5), and Conclusions and future perspectives (Chapter 6). The results chapters are in the form of PDFs of studies already published in peer-reviewed journals or manuscripts formatted to meet the requirements of the journals to which they have been submitted. This thesis has been prepared in accordance with the Griffith University policy on preparing a PhD thesis as a series of published and unpublished papers (Appendix 1). As a result, there is some repetition among the results chapters, including the descriptions of study methods and detailed literature reviews in accordance with the requirements of each journal. The manuscripts presented in this thesis are as follow:

1.6.1 Publications


1.6.2 Publications under review

Jianyin Huang, William W. Bennett, David T. Welsh and Peter R. Teasdale. Diffusive gradients in thin films techniques provide representative time-weighted average measurements of inorganic nutrients in dynamic freshwater systems. *Environmental Science and Technology* (Chapter 5).

1.6.3 Additional relevant publication in preparation

Jianyin Huang, William W. Bennett, Peter R. Teasdale, David T. Welsh. Removing ammonium from water and wastewater using emerging adsorbents – A review. *Chemical Engineering Journal*.

1.7 References


55. EPA; WSAs *European Communities (Drinking Water) (No. 2) Regulations* Environmental Protection Agency & Water Service Authorities for Public Water Supplies 2007


2. Development and evaluation of a diffusive gradients in thin films technique for measuring nitrate in freshwaters

The introduction chapter identified the problem of increasing nitrate concentrations in freshwaters and the issues associated with using grab water sampling techniques to monitor nitrate concentrations within the aquatic systems. This chapter addresses the first aim of the thesis, to develop a diffusive gradients in thin films (DGT) technique using A520E anion exchange resin for monitoring nitrate concentrations in freshwaters. The performance of A520E-DGT was systematically investigated. This includes determination of the uptake and elution efficiencies, demonstration of linear mass accumulation over time, evaluation of the effects of various pH and ionic strengths, and individual competing anions, measurement of the intrinsic binding capacity, evaluation of the performance in a synthetic freshwater matrix, and validation of A520E-DGT methods at two field sites – Loders Creek and Saltwater Creek, on the Gold Coast, Australia.

This chapter has been published at Analytica Chimica Acta and the information of this paper was provided.

The co-authors of this manuscript are my thesis supervisors, Prof. Peter Teasdale, Assoc. Prof. David Welsh and Dr. William Bennett. They provided feedback on the experimental design and manuscript drafts. Honours student Sean Gardiner provided help with field experiments. My (Jianyin Huang) contribution to the manuscript involved: developing the experimental design, all data collection and analysis, and the preparation of the manuscript.
Jianyin Huang

Corresponding Author: Prof. Peter Teasdale

Principal Supervisor: Prof. Peter Teasdale

Principal Supervisor: Assoc. Prof. David Welsh
Development and evaluation of the diffusive gradients in thin films technique for measuring nitrate in freshwaters

Jianyin Huang, William W. Bennett, Peter R. Teasdale*, Sean Gardiner, David T. Welsh

Environmental Futures Research Institute, Griffith University, Gold Coast Campus, QLD 4222, Australia

HIGHLIGHTS

- A new DGT technique, using Purolite A520E was developed and evaluated for monitoring NO$_3$-N in freshwaters.
- A520E-DGT is suitable for a wide range of pH from 3.5 to 8.5 and ionic strengths up to 0.008 mol L$^{-1}$ NaCl.
- A520E-DGT showed a good linear uptake for NO$_3$-N over 72 h under high NO$_3$-N concentration.
- During field deployments, Caco$_2$ values with DBL matched excellently with the average grab sample values.

ABSTRACT

A new diffusive gradients in thin films (DGT) technique, using Purolite A520E anion exchange resin, was developed and evaluated for the measurement of NO$_3$-N in freshwaters. Purolite A520E had a very high uptake efficiency (>98%) and elution efficiency (82.7% with 2 mol L$^{-1}$ NaCl as eluent) for NO$_3$-N. The 24 h mass vs. time validation experiments had excellent linearity ($R^2 > 0.997$) and the intrinsic capacity of the binding layer for NO$_3$-N was 849 ± 24 μg NO$_3$-N uptake over a pH (3.5–8.5) range typical of most natural freshwaters. Several anions competed with NO$_3$-N to produce a lower effective binding capacity for NO$_3$-N in the following order of selectivity, CI$^-$ > HCO$_3$$^-$ > SO$_4^{2-}$ > HPO$_4^{2-}$, although NO$_3$-N measurements were quantitative at ionic strengths 0.0001–0.008 mol L$^{-1}$ as NaCl. NO$_3$-N did not adversely affect determination of NO$_2$-N at typical concentrations. Field deployments of DGT samplers with varying diffusive layer thicknesses validated the use of the technique in situ, allowing calculation of the diffusive boundary layer and accurate measurement of NO$_3$-N (Caco$_2$/CDBL 1.03–1.04). Reproducibility of the technique during field deployments was good (relative standard deviation < 3.2%). Limits of detection of A520E-DGT for NO$_3$-N were 13.4 μg L$^{-1}$ and 7.52 μg L$^{-1}$ (equivalent to 0.94 and 0.54 μmol L$^{-1}$) based on 24 h and 48 h deployments, respectively. A520E-DGT determined NO$_3$-N concentrations during field deployments were very similar to the average values obtained from 0.45 μm filtered grab samples, which confirmed that the new DGT technique produced highly representative results.

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3. Development and evaluation of a diffusive gradients in a thin film technique for measuring ammonium in freshwaters

The introduction chapter presented the problem of increasing ammonium levels in freshwaters and the disadvantages of using grab sampling techniques to monitor ammonium concentrations in the aquatic systems. This second results chapter is related to the second aim of the thesis to develop a diffusive gradients in thin films (DGT) technique using PrCH cation exchange resin to monitor ammonium concentrations in freshwaters. The chapter provides laboratory and field evaluations of PrCH-DGT, including determination of the uptake and elution efficiencies, evaluation of the influences of different ionic strengths on the effective diffusion coefficient, evaluation of the effects of various pH and potential interfering cations, measurement of the intrinsic binding capacity, validation of the PrCH-DGT method in synthetic freshwater and at two field sites – Mudgeeraba Creek and Loders Creek, on the Gold Coast, Australia.

This chapter has been published at Analytica Chimica Acta and the information of this paper was provided.

The co-authors of this manuscript are my thesis supervisors, Prof. Peter Teasdale, Assoc. Prof. David Welsh and Dr. William Bennett. They provided feedback on the experimental design, interpretation of the data and manuscript drafts. PhD student Tianling Li provided help with field experiments. My (Jianyin Huang) contribution to the manuscript involved: devising the experimental design, all data collection and analysis, and preparation of the manuscript.
Jianyin Huang

Corresponding Author: Assoc. Prof. David Welsh

Principal Supervisor: Prof. Peter Teasdale

Principal Supervisor: Assoc. Prof. David Welsh
Development and evaluation of a diffusive gradients in a thin film technique for measuring ammonium in freshwaters

Jianyin Huang, William W. Bennett, David T. Welsh*, Tianling Li, Peter R. Teasdale

Environmental Futures Research Institute, School of Environment, Griffith University, Gold Coast Campus, QLD 4222, Australia

HIGHLIGHTS

- A new DGT technique, using Micro-lite PrOH, was developed and evaluated for monitoring NH$_4^+$-N in freshwaters.
- PrOH-DGT is suitable for a wide range of pH conditions (3.5–8.5) and ionic strengths (up to 0.012 mol L$^{-1}$ NaCl).
- PrOH-DGT showed a good linear uptake for NH$_4^+$-N over 72 h in synthetic freshwater.
- During field deployments, C$_{DGT}$ values with DBL matched well with the average grab sample values.

ARTICLE INFO

Article History:
Received 25 September 2015
Received in revised form
11 November 2015
Accepted 16 November 2015
Available online 30 November 2015

Keywords:
Passive sampler
Nitrogen
Ammonium diffusion coefficient
Diffusive boundary layer
Variability
Freshwater streams

ABSTRACT

A new diffusive gradients in a thin film (DGT) technique, using Microlite PrOH cation exchange resin, was developed and evaluated for measuring NH$_4^+$-N in freshwaters. Microlite PrOH had high uptake (>92.5%) and elution efficiencies (87.2% using 2 mol L$^{-1}$ NaCl). Mass vs. time validation experiments over 24 h demonstrated excellent linearity ($R^2 = 0.996$). PrOH-DGT binding layers had an extremely high intrinsic binding capacity for NH$_4^+$-N (~3000 µg). NH$_4^+$-N uptake was quantitative over pH ranges 3.5–8.5 and ionic strength (up to 0.012 mol L$^{-1}$ as NaCl) typical of freshwater systems. Several cations (Na$^+$, K$^+$, Ca$^{2+}$ and Mg$^{2+}$) were found to compete with NH$_4^+$-N for uptake by PrOH-DGT, but NH$_4^+$-N uptake was quantitative over concentration ranges typical of freshwater (up to 0.012 mol L$^{-1}$ Na$^+$, 0.006 mol L$^{-1}$ K$^+$, 0.003 mol L$^{-1}$ Ca$^{2+}$ and 0.004 mol L$^{-1}$ Mg$^{2+}$). Effective diffusion coefficients determined from mass vs. time experiments changed non-linearly with electrical conductivity. Field deployments of DGT samplers with varying diffusive layer thicknesses validated the use of the technique in situ, allowing deployment times to be manipulated with respect to NH$_4^+$-N concentration, and enable the calculation of the diffusive boundary layer thickness. Daily grab sample NH$_4^+$-N concentrations were observed to vary considerably independent of major rainfall events, but good agreements were obtained between PrOH-DGT values and mean grab sample measurements of NH$_4^+$-N ($C_{DGT}/C_{GRAB}$ 0.83–1.3). Reproducibility of DGT measurements in the field was good (relative standard deviation < 15%). Limit of detection was 0.03 µg L$^{-1}$ (equivalent to 0.045 µmol L$^{-1}$) based on 24 h deployments.

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4. Determining time-weighted average concentrations of nitrate and ammonium in freshwaters using DGT with ion exchange membrane-based binding layers

The preceding chapters described the development of DGT techniques for monitoring nitrate and ammonium using traditional binding hydrogels incorporating analyte specific ion exchange resins. This third results chapter investigates alternative methods for measuring nitrate and ammonium using commercially available AMI-DGT and CMI-DGT anion and cation exchange membranes as the DGT binding layers. The performances of the AMI-DGT and CMI-DGT methods were systematically evaluated in laboratory and field experiments, and directly compared with that of A520E-DGT and PrCH-DGT, respectively.

This chapter is currently under review for *Environmental Science: Processes & Impacts* and has been formatted in the journal style.

The co-authors of this manuscript are my thesis supervisors, Prof. Peter Teasdale, Assoc. Prof. David Welsh and Dr. William Bennett. They provided feedback on experimental design and manuscript drafts. My (Jianyin Huang) contribution to the manuscript involved: developing the experimental design, all data collection and analysis, and preparation of the manuscript.
Jianyin Huang

Corresponding Author: Assoc. Prof. David Welsh

Principal Supervisor: Prof. Peter Teasdale

Principal Supervisor: Assoc. Prof. David Welsh
Determining time-weighted average concentrations of nitrate and ammonium in freshwaters using DGT with ion exchange membrane-based binding layers

Jianyin Huang, William W. Bennett, David T. Welsh and Peter R. Teasdale

Abstract. Commercially-available AMI-7001 anion exchange and CMI-7000 cation exchange membranes were utilised as binding layers for DGT measurements of NO$_3$-N and NH$_4$-N in freshwaters. These ion exchange membranes are easier to prepare and handle than DGT binding layers consisting of hydrogels cast with ion exchange resins. The membranes showed good uptake and elution efficiencies for both NO$_3$-N and NH$_4$-N. The membrane-based DGTs are suitable for pH 3.5 - 8.5 and ionic strength ranges (0.0001 - 0.014 and 0.0003 - 0.012 mol L$^{-1}$ as NaCl for the AMI-7001 and CMI-7000 membrane, respectively) typical of most natural freshwaters. The binding membranes had high intrinsic binding capacities for NO$_3$-N and NH$_4$-N of 911 ± 88 μg and 3512 ± 51 μg, respectively. Interferences from the major competing ions for membrane-based DGTs are similar to DGTs employing resin-based binding layers but with slightly different selectivity. This different selectivity means that the two DGT types can be used in different types of freshwaters. The laboratory and field experiments demonstrated that AMI-DGT and CMI-DGT can be an alternative to A520E-DGT and PrCH-DGT for measuring NO$_3$-N and NH$_4$-N, respectively, as (i) membrane-based DGT have a consistent composition, (ii) avoid the use of toxic chemicals, (iii) provided highly representative results with $C_{\text{DGT}}:C_{\text{GDL}}$ between 0.81 and 1.3, and (iv) provided similar results to resin-based DGTs with $C_{\text{membrane-DGT}}:C_{\text{resin-DGT}}$ between 0.85 and 1.2 during the deployment time.

1. Introduction

Nitrate (NO$_3$-N) and ammonium (NH$_4$-N) are highly bioavailable forms of nitrogen that are important nutrients for primary producers like algae and aquatic vegetation. The concentrations of NO$_3$-N and NH$_4$-N are generally low in natural waters, however, they can reach very high levels due to the widespread use of fertilizers and human sewage inputs. Nitrogen pollutions have led to widespread ecological degradation, algal blooms and hypoxic events in freshwater bodies and estuaries around the world. Additionally, high concentrations of NO$_3$-N and NH$_4$-N in drinking water are associated with blue baby syndrome and NH$_4$ can be toxic to fish. There is, therefore, a need to monitor NO$_3$-N and NH$_4$-N concentrations in freshwater bodies to maintain their ecological values and protect potable water supplies.

The most common methods for determining NO$_3$-N and NH$_4$-N concentrations in waterbodies involve the instrumental analysis of grab samples. These methods, however, are unlikely to provide representative data for nutrient concentrations due to the low frequency of sampling and the high variability in concentrations that can occur with changes in weather, land uses, inputs from point and non-point sources and the influences of diurnal and seasonal cycles on N-cycling within the water bodies. Therefore, alternative techniques capable of providing more representative measurements are required. These include passive sampling techniques, which integrate short-term variations in analyze concentrations and provide time-weighted average concentrations over environmentally relevant time-scales.

The diffusive gradients in thin films (DGT) technique is a well-established passive sampling technique for measuring a range of elemental contaminants. The standard DGT device consists of a circular cap with a 2.00-cm-diameter open window, a protective filter membrane overlaying a diffusive layer of known thickness (Δg), a binding layer and a piston. Both layers are commonly polyacrylamide hydrogels, with the
binding layer containing a resin or substance selective for the analyte species. A range of binding agents have been used for DGT techniques: the chelating resin Chelex-100 for Cu, Zn, Ni, etc.,22, 23 various metal oxide materials for oxyanions, including ferrihydrite,24 the TiO₂-based adsorbent (Metsorb),16, 20 and zirconium oxide,25 and ion exchange materials, such as AS20E resin for NO₃⁻N,18 and PrCH resin19 and zeolite26 for NH₄-N. The preparation of consistent and high-quality binding layers can be time-consuming and difficult for inexperienced users, which can lead to measurement errors. Li et al. described a cellulose phosphate cation exchange membrane as a DGT binding layer for measuring trace metals.27 In contrast to laboratory-made binding resin gels, membranes- or paper-based binding layers are commercially manufactured with a consistent composition,28 which make them highly suitable for routine monitoring applications of DGT where quality control and ease of use are paramount.

In recent decades, ion exchange membranes have been developed from laboratory tools to industrial products with significant technical and commercial impacts.29 They have been widely used for desalination of seawater, wastewater treatment, and concentration and separation of pharmaceutical and food products.29 Anion exchange membranes contain positively charged groups, including –NH₃⁺, –NRH₂⁺, –NR₂H⁺ and –SR⁺, while cation exchange membranes contain negatively charged groups, including –SO₃⁻, –COO⁻, –PO₃²⁻ and C₂H₄O₂⁻. AMI-7001 anion exchange membranes and CMI-7000 cation exchange membranes have been utilised in microbial fuel cells and desalination.30, 31

In this study, AMI-7001 anion exchange membranes consisting of quaternary ammonium functional groups (–NRH₂⁺, CI form) and CMI-7000 cation exchange membranes with sulfonic acid functional groups (–SO₃⁻, Na⁺ form) were selected for evaluation in the DGT measurement of NO₃⁻N and NH₄-N, because they use the same functional groups as two ion exchange resins recently described for use with DGTs.18, 19 Performances of AMI-7001 and CMI-7000 ion exchange membranes as new binding layers for DGT measurements of NO₃⁻N and NH₄-N, respectively, were systematically investigated. These included determination of their uptake and elution efficiencies, demonstration of linear mass accumulation over time, evaluation of the effects of various pH and ionic strengths, measurement of their intrinsic binding capacities, and evaluation of their performances in a synthetic freshwater matrix. The performance of the membrane-based AMI-DGT was compared with AS20E-DGT for NO₃⁻N18 and the performance of CMI-DGT was compared with PrCH-DGT for NH₄-N.19 Finally, the two membrane- and resin-based binding layer DGT techniques were directly compared in field deployment and with NO₃⁻N and NH₄-N concentrations obtained from time-series grab sampling.

2. Experimental

2.1 General methods and materials

Deionised water (Milli-Q Advantage A10, 18.2 MΩ) was used to prepare all solutions, reagents and for all washing. All chemicals were analytical reagent grade or higher. Nitrate and ammonium solutions were prepared from standard solutions of 1000 mg L⁻¹ nitrogen as NaNO₃ (AR grade, Merck) and NH₄Cl (AR grade, Merck), respectively. AMI-7001 anion exchange membranes and CMI-7000 cation exchange membranes (both 0.45 ± 0.025 mm) were purchased from Membranes International Inc. USA. 0.45 mm polysulfone filter membrane was provided by VWR Australia. The plastic containers used for the preparation and storage of solutions and experimental work, glass plates used for preparing diffusive and binding gels, and DGT probe components, were washed in 10% (v/v) HCl (AR grade, Merck) for 24 h and rinsed thoroughly with deionised water prior to use.

2.2 Preparation of hydrogel-based diffusive and binding layers

Agarose-cross-linked polyacrylamide diffusive layers were prepared according to Zhang and Davison.21 Agarose diffusive layers were prepared as described previously by Huang et al.19 Diffusive gels were stored in 0.001 mol L⁻¹ NaCl solution prior to assembly in the DGT samplers. AS20E and PrCH resin-based binding layers were prepared as described by Huang et al.18, 19 The detailed procedures were presented as follow: for preparing AS20E binding layer, 2 g of fine AS20E powder was added to 4 mL bisacrylamide gel stock solution. 400 µL of ammonium persulphate (Chem-Supply Pty. Ltd.) and 95 µL of N,N,N’,N’-tetramethyl ethylenediamine (TEMED, Merck) were added, the solutions were mixed well for 1 min on a magnetic stirrer and the gels cast between two glass plates separated by inert 0.50 mm spacers. For preparing PrCH binding layer, 2.5 g of PrCH was added and mixed into 6 mL of agarose solution prior to casting. The glass plates used for casting were separated by 0.75 mm thick spacers. Both binding layers were set for 45 minutes at room temperature, then washed 2 - 3 times before being stored in deionised water at 4 °C.

2.3 Preparation of membrane binding layers

AMI-7001 anion exchange membranes and CMI-7000 cation exchange membranes were pre-conditioned by immersion in a 5% NaCl solution at 40 °C for 24 h to allow membrane hydration and expansion. The membranes were then washed with deionised water at least three times to remove residual NaCl, cut into 4.91 cm circles and stored in deionised water. Polysulphone membranes (0.45 µm, VWR) were soaked in deionised water for 24 h, washed in 5% (v/v) HCl for a further 24 h, thoroughly rinsed with deionised water and stored in 0.001 mol L⁻¹ NaCl prior to use.

2.4 DGT assembly and DGT theory

DGT samplers (purchased from DGT Research Ltd) with an open window area of 3.14 cm² were assembled by overlaying the various binding layers with a diffusive gel disc and a
polysulfone filter membrane, as described previously.\textsuperscript{18, 19} Assembled bags were stored at 4 °C in sealed double plastic bags containing 1 - 2 mL of deionised water.

The analyte concentration in solution ($C_{\text{DGT}}$, ng mL\textsuperscript{-1}, which is equivalent to μg L\textsuperscript{-1}) is calculated using Fick's first law of diffusion and the measured mass of the analyte accumulated on the binding layer over a known deployment time.$^{21, 22}$

\[
C_{\text{DGT}} = \frac{M\Delta g}{DAt}
\]  

\(M\) is the mass of the analyte species bound to the binding phase (ng) corrected for the elution efficiency, \(\Delta g\) is the diffusive layer thickness (cm), \(D\) is the diffusion coefficient of the analyte species through the diffusive layer (cm\textsuperscript{2} s\textsuperscript{-1}), \(t\) is the deployment time (s) and \(A\) is the area of the probe exposed to solution (cm\textsuperscript{2}). The NO\textsubscript{3}\textsuperscript{-} diffusion coefficient of $1.46 \times 10^{-5}$ cm\textsuperscript{2} s\textsuperscript{-1} at 25 °C from Huang \textit{et al.} was used for all calculations and corrected for temperature as appropriate using the Stokes-Einstein equation.$^{18, 22}$ The determination of the diffusion coefficient is more complex for NH\textsubscript{4}+, especially at low ionic strengths.$^{19}$ Therefore, experiments were conducted to better understand the effects of solution ionic strengths on the diffusion coefficient of NH\textsubscript{4}+ (see section 2.6.2). These diffusion coefficients were also corrected for temperature.

2.5 Nitrate and ammonium analyses

NO\textsubscript{3}-N and NH\textsubscript{4}-N were measured using automated colorimetric methods (APHA Standard Methods 4500- NO\textsubscript{3} F and 4500- NH\textsubscript{3} H) using a Seal AA3 segmented flow analyser. Analytical standards were prepared from 1000 mg L\textsuperscript{-1} NO\textsubscript{3} and NH\textsubscript{4}+ standard solutions (AR grade, Merck). Quality control standards were prepared using dried potassium nitrate (KNO\textsubscript{3}) and ammonium sulfate ((NH\textsubscript{4})\textsubscript{2}SO\textsubscript{4}) and analysed every 23 samples. The instrumental detection limits for NO\textsubscript{3}-N and NH\textsubscript{4}-N were 1.4 μg L\textsuperscript{-1} and 1.25 μg L\textsuperscript{-1}, respectively (determined as 3 \times the standard deviation of the blanks, \(n = 10\)).

2.6 Laboratory evaluation

2.6.1 Uptake and elution efficiencies

Uptake and elution efficiencies were measured by immersing triplicate AMI-7001 and CMI-7000 ion exchange membrane discs in triplicate 10 mL of NO\textsubscript{3}-N solutions (0.11 mg L\textsuperscript{-1} to 4.5 mg L\textsuperscript{-1}) and NH\textsubscript{4}-N solutions (0.156 mg L\textsuperscript{-1} to 7.78 mg L\textsuperscript{-1}) at pH 7.0 \pm 0.5, respectively. After 24 h the membrane discs were removed and eluted in 2 mol L\textsuperscript{-1} NaCl solution for 24 h. As AMI and CMI had the same functional groups as two recently described DGT techniques,$^{18, 19}$ the same eluent (2 mL of 2 mol L\textsuperscript{-1} NaCl) was used for this study. Samples of the deployment solutions were analysed to determine the mass of analyte remaining in solution. The uptake efficiency was determined as the percentage of NO\textsubscript{3}-N or NH\textsubscript{4}-N removed from the solution and the elution efficiency was the percentage of the mass in the eluent over the mass removed.

2.6.2 Effect of ionic strength on the NH\textsubscript{4}+ diffusion coefficient

To investigate the effects of solution ionic strengths on the diffusion coefficient of NH\textsubscript{4}+, CMI-DGT NH\textsubscript{4}-N mass accumulation over 24 h was conducted in synthetic freshwaters with different targeted ionic strengths (0.080, 0.20, 0.40, 0.80 and 1 mS cm\textsuperscript{-1}) and constant NH\textsubscript{4}-N (1.5 \pm 0.21 mg L\textsuperscript{-1}) concentration. Five sets of triplicate CMI-DGT samplers were deployed in each synthetic freshwater, which was prepared based on modified Langmuir \textit{et al.}\textsuperscript{23} as previously described.$^{19}$ The diffusion coefficient of NH\textsubscript{4}+ was determined from DGT mass accumulation over time using Eq. 2.$^{22}$

\[
D = \frac{a\Delta g}{CA}
\]

Where \(a\) is the slope of the linear regression of the mass of analyte (ng) accumulated in the binding gel over time (s), \(\Delta g\) is the thickness of the diffusive layer (polyacrylamide diffusive gel and filter membrane) (cm), \(A\) is the area of the diffusive layer available for diffusion (cm\textsuperscript{2}) (3.14 cm\textsuperscript{2} was used for laboratory experiment in well-stirred solutions and 3.8 cm\textsuperscript{2} was used in the field where DBLs were present)$^{33}$ and \(C\) is the concentration of analyte in the solution (ng mL\textsuperscript{-1}).

2.6.3 Effect of pH and ionic strength on uptake

The effects of pH and ionic strengths on the accumulation of NO\textsubscript{3}-N and NH\textsubscript{4}-N were investigated by deploying triplicate AMI-DGTs and CMI-DGTs in mixed 7 L solutions of NO\textsubscript{3}-N (1.0 mg L\textsuperscript{-1}) and NH\textsubscript{4}-N (1.5 mg L\textsuperscript{-1}) at different pH (3.5, 5.0, 7.0 and 8.5 with 0.002 mol L\textsuperscript{-1} NaCl) and various ionic strengths (0.0001, 0.001, 0.01 and 0.1 mol L\textsuperscript{-1} NaCl with pH 7.0 \pm 0.5) for 24 h. 1 mol L\textsuperscript{-1} NaOH and 1 mol L\textsuperscript{-1} HCl solutions were used to adjust the solution pH. Solutions were equilibrated for 24 h prior to the deployment of DGT samplers. Grab samples of the solutions were collected for NO\textsubscript{3}-N and NH\textsubscript{4}-N analyses at the beginning of each experiment and after the removal of DGT samplers.

2.6.4 Effect of competing ions

The effects of potentially competing anions (Cl\textsuperscript{-}, HCO\textsubscript{3}\textsuperscript{-} and SO\textsubscript{4}\textsuperscript{2-}) on the measurement of NO\textsubscript{3}-N by AS20E-DGT and AMI-DGT were investigated by time-series deployment of triplicate DGT samplers in NO\textsubscript{3}-N solutions (0.9 mg L\textsuperscript{-1}) with different anion concentrations: Cl\textsuperscript{-} (0.0005, 0.005, 0.012 mol L\textsuperscript{-1}) as NaCl; HCO\textsubscript{3}\textsuperscript{-} (0.0005, 0.005, 0.012 mol L\textsuperscript{-1}) as NaHCO\textsubscript{3}; and SO\textsubscript{4}\textsuperscript{2-} (0.0005, 0.0025, 0.005 mol L\textsuperscript{-1}) as Na\textsubscript{2}SO\textsubscript{4}. The effects of competing cations (Na\textsuperscript{+}, K\textsuperscript{+}, Ca\textsuperscript{2+} and Mg\textsuperscript{2+}) on NH\textsubscript{4}-N uptake of PrCH-DGT and CMI-DGT were investigated in the same manner by deploying DGT samplers in NH\textsubscript{4}-N solutions (0.6 mg L\textsuperscript{-1}) with differing cation concentrations: Na\textsuperscript{+} (0.0005, 0.002, 0.005, 0.01 mol L\textsuperscript{-1}) as NaCl; K\textsuperscript{+} (0.0005, 0.002, 0.005, 0.01 mol L\textsuperscript{-1}) as KCl; Ca\textsuperscript{2+} (0.0003, 0.0012, 0.0025 and 0.005 mol L\textsuperscript{-1}) as CaCl\textsubscript{2}; H\textsubscript{2}O; and Mg\textsuperscript{2+} (0.0003, 0.0015, 0.003 and 0.006 mol L\textsuperscript{-1}) as MgCl\textsubscript{2}.6H\textsubscript{2}O. The pH of each solution with different
concentrations of anions and cations were recorded and summarised in the caption of Figures 1 and 2. The selected concentrations of each anion and cation, except the lowest concentration, are well above those found in most surface freshwater around the world.\textsuperscript{14}

2.6.5 Evaluation of the AMI binding capacity for NO$_3$-N and CMI binding capacity for NH$_4$-N

The intrinsic NO$_3$-N and NH$_4$-N binding capacities of AMI-7001 and CMI-7000 membranes were determined by deploying nine sets of triplicate AMI-DGT and CMI-DGT samplers in 11.29 mg L$^{-1}$ NO$_3$-N and 30 mg L$^{-1}$ NH$_4$-N solutions without any competing ions for 72 h. A set of DGT samplers was removed after 4 h, 8 h and then every 8 h up to 72 h. Samples of the solutions (5 mL) were taken at the beginning of the experiment and after the removal of each set of DGT samplers with 10 samples collected in total.

2.6.6 Evaluation of performance in synthetic freshwater

Seven sets of triplicate DGT samplers of each type with $\Delta g$ of 0.09 cm were deployed in synthetic freshwater (prepared as per\textsuperscript{35}) with relatively low concentrations of NO$_3$-N (0.3 mg L$^{-1}$) and NH$_4$-N (0.1 mg L$^{-1}$) to evaluate their performance. Triplicate DGT samplers were retrieved for analysis after 4, 8, 12, 24, 48 and 72 h, and grab samples of analyte solutions were collected prior to deployment and after the removal of each set of DGT samplers.

3. Field performance in natural freshwaters

Four sets of DGT samplers of each type were deployed at two field sites on the Gold Coast, Queensland, Australia. The first site was Loders Creek, a small urban stream with a catchment of approximately 10 km$^2$, dominated by urban and industrial land uses.\textsuperscript{35, 36} The second site was Saltwater Creek, a small micro-tidal estuary, flowing from its headwaters in Nerang State Forest to the Coombabah Creek estuary confluence.\textsuperscript{33} The creek system is approximately 17 km long, with the catchment being 13.5% urban and 36% grazing and cultivation.

DGT samplers of each type were deployed in triplicate at a water depth of approximately 30 cm for periods of approximately three days over nine consecutive days. Additional DGT samplers with various diffusive layer thicknesses ($\Delta g$) (0.05 - 0.13 cm) were deployed to calculate the thickness of the diffusive boundary layer (DBL) as described by Zhang and Davison.\textsuperscript{37} At the completion of the deployment, DGT samplers were briefly rinsed with deionised water, placed in acid-washed plastic bags and stored at $< 4 \, ^\circ\text{C}$ until analysis. Water temperature, salinity/conductivity and pH were recorded daily using a calibrated combination meter (TPS 90-FLMV) and 0.45 $\mu$m filterable grab water samples for NO$_3$-N and NH$_4$-N analyses were collected daily over the deployment time.

3. Results and discussion

3.1 Uptake and elution efficiencies

The uptake efficiencies of the AMI-7001 and CMI-7000 binding membranes for NO$_3$-N and NH$_4$-N were 96.7 ± 3.3% and 94.8 ± 2.3%, respectively, over the studied mass ranges of NO$_3$-N (1.13 - 45.16 $\mu$g) and NH$_4$-N (1.56 - 77.78 $\mu$g), demonstrating quantitative uptake within 24 h. The elution efficiency of NO$_3$-N from the AMI-7001 membrane was 77.6 ± 6.1% and the elution efficiency of NH$_4$-N from CMI-7000 was 89.9 ± 4.6%, over the tested mass range. Both uptake and elution efficiencies are similar to those of NO$_3$-N from A520E binding gels\textsuperscript{18} and NH$_4$-N from PrCh binding gels\textsuperscript{19} (Supplementary Information, Table S1), and are consistent with those reported for other DGT techniques.\textsuperscript{16, 17, 22} The low standard deviations of the elution efficiencies indicate that the elution procedures were sufficiently reproducible.

3.2 Effect of solution ionic strength (conductivity) on NH$_4$\textsuperscript{+} diffusion coefficient

Diffusion coefficients ($D$, cm$^{2}$ s$^{-1}$) of NH$_4$\textsuperscript{+} were determined from DGT mass accumulation over time experiments in synthetic freshwaters with a range of conductivity values. The results (Figures S1 and S2) showed that NH$_4$\textsuperscript{+} diffusion coefficients increased with decreasing solution conductivity as was previously observed for PrCh-DGT\textsuperscript{19} (data and curve were also shown in Figure S2), which is consistent with the diffusion properties being independent of the binding layer. The trend lines of diffusion coefficients with conductivity were modelled using regression analysis - exponential curves provided an excellent and highly significant fit to the data ($R^2 = 0.9979$ and p < 0.01) for CMI-DGT. The equation used to predict $D$ for a given conductivity value was $D = 0.000115 \times \text{Conductivity}^{0.346833}$ which was then modified for temperature using the Stokes-Einstein equation.\textsuperscript{22}

The increase in the determined diffusion coefficients at low ionic strength may be attributed to the presence of negative charges (carboxylic functional groups) within the agarose diffusive gel, as reported in previous studies,\textsuperscript{19, 38} which results in enhanced or retarded diffusion for which the effects are greater at low ionic strength. The difference between the two curves is low at conductivity $> 500 \, \mu$S cm$^{-1}$, with an uncertainty of about 10%. The uncertainty increases to about 20% at conductivity $< 200 \, \mu$S cm$^{-1}$. Variation of $D$ for NH$_4$\textsuperscript{+} with conductivity was also observed in a recent study\textsuperscript{26} in which the normal polyacrylamide hydrogel was used, although the effect was less dramatic than observed here. We did not use a polycrylamide diffusive or binding layer because of concerns over interference from the amide functional groups with analysis of NH$_4$-N, although this was not reported to be an issue.\textsuperscript{26}

3.3 Effect of pH and ionic strength on DGT measurement

The ratios of $C_{\text{AMI-DGT:CSOLN}}$ and $C_{\text{CMI-DGT:CSOLN}}$ for pH solutions 3.5, 5.0, 7.0, and 8.5 were between 0.87 and 1.06, indicating reasonable performance as the acceptable range is 0.9 - 1.1. The results were similar to those of previous DGT methods.
(A520E-DGT and PrCH-DGT) for NO$_3$-N$^{18}$ and NH$_4$-N$^{18}$ (Table S2). The independent t-tests indicated that there was no significant difference ($p > 0.05$) between the resin-based binding layers and the membrane binding layers at different pH, although the ratios of $C_{\text{AMI-DGT}}$/$C_{\text{SOLN}}$ and $C_{\text{CMI-DGT}}$/$C_{\text{SOLN}}$ in acidic solutions was slightly lower at pH 3.5.

The effects of solution ionic strengths on DGT measurements of NO$_3$-N (Table 1) and NH$_4$-N (Table 2) were evaluated and the performances of the membrane and resin-based binding layers were compared. The $C_{\text{DGT}}$/$C_{\text{SOLN}}$ values for NO$_3$-N were close to 1 at ionic strengths of 0.0001 and 0.001 mol L$^{-1}$ NaCl for both AMI- and A520E-DGT over 24 h deployments. At ionic strengths of 0.01 and 0.1 mol L$^{-1}$ NaCl, $C_{\text{DGT}}$/$C_{\text{SOLN}}$ decreased to 0.85 and 0.2 for AMI-DGT, and 0.72 and 0.1 for A520E-DGT, respectively, suggesting that NO$_3$-N measurements by A520E-DGT were slightly more sensitive to ionic strength than those by AMI-DGT. For NH$_4$-N, $C_{\text{DGT}}$/$C_{\text{SOLN}}$ values were close to 1 for both CMI- and PrCH-DGT at ionic strengths from 0.0001 to 0.01 mol L$^{-1}$ NaCl, but both DGT techniques grossly underestimated solution concentrations at 0.1 mol L$^{-1}$ NaCl over 24 h deployments. Further investigations (data not shown) determined that AMI-DGT accurately determined NO$_3$-N up to 0.014 mol L$^{-1}$ NaCl (conductivity = 1.4 mS cm$^{-1}$) and CMI-DGT accurately measured NH$_4$-N up to 0.012 mol L$^{-1}$ NaCl (conductivity = 1.2 mS cm$^{-1}$) over 24 h deployments. AMI-DGT had wider ionic strength range compared with A520E-DGT (up to 0.008 mol L$^{-1}$ NaCl) and CMI-DGT had the same ionic strength range as PrCH-DGT (up to 0.012 mol L$^{-1}$ NaCl).

The wide range of conductivities over which the new DGTs accurately measured NO$_3$-N and NH$_4$-N concentrations recommend that the AMI-DGT and CMI-DGT methods are suitable for determining nutrient concentrations in most freshwaters within short periods, as these are defined as having a conductivity of lower than 1.5 mS cm$^{-1}$ in the Australian Freshwater Quality Guidelines. We propose that in freshwaters with conductivity ≤ 1 mS cm$^{-1}$ both techniques are suitable for determination of their respective analyte. If conductivity is > 1 mS cm$^{-1}$, the performance of the two DGT methods should be further investigated at the specific major anion/cation concentration levels of the waterway.

Table 1. Comparison of the DGT measurements for NO$_3$-N at different ionic strengths (mol L$^{-1}$ NaCl). Data are mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Ionic strength</th>
<th>$C_{\text{AMI-DGT}}$/$C_{\text{SOLN}}$</th>
<th>$C_{\text{A520E-DGT}}$/$C_{\text{SOLN}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0001</td>
<td>1.10 ± 0.06</td>
<td>1.02 ± 0.05</td>
</tr>
<tr>
<td>0.001</td>
<td>0.92 ± 0.09</td>
<td>0.90 ± 0.02</td>
</tr>
<tr>
<td>0.01</td>
<td>0.85 ± 0.17</td>
<td>0.72 ± 0.03</td>
</tr>
<tr>
<td>0.1</td>
<td>0.20 ± 0.02</td>
<td>0.10 ± 0.001</td>
</tr>
</tbody>
</table>

3.4 Anionic interferences with NO$_3$-N measurements by AMI-DGT

As the effects of competition become more substantial for longer deployments, the effects of Cl$^-$, HCO$_3^-$ and SO$_4^{2-}$ on NO$_3$-N measurements by AMI-DGT were investigated and compared with A520E-DGT by deploying DGT samplers of each type for 72 h in solutions with varying anion concentrations (Figure 1). $C_{\text{DGT}}$/$C_{\text{SOLN}}$ values were between 0.85 and 1 for AMI-DGT and A520E-DGT at 0.0005 and 0.005 mol L$^{-1}$ Cl$^-$ and HCO$_3^-$, and 0.0005 and 0.0025 mol L$^{-1}$ SO$_4^{2-}$. At 0.012 mol L$^{-1}$ Cl$^-$ and HCO$_3^-$, and 0.005 mol L$^{-1}$ SO$_4^{2-}$, $C_{\text{DGT}}$/$C_{\text{SOLN}}$ ratios became unacceptable at ~0.6 or lower (ratios < 0.8 are considered to be inaccurate). These results indicate that the interferences of different competing ions are slightly different for the selected binding layers. The affinity of the adsorbents for the tested anions decreased in the order SO$_4^{2-}$ > HCO$_3^-$ > Cl$^-$ for AMI-DGT and Cl$^-$ > HCO$_3^-$ > SO$_4^{2-}$ for A520E-DGT. Both DGT methods can accurately measure NO$_3$-N up to 0.005 mol L$^{-1}$ Cl$^-$ (178 mg L$^{-1}$) and HCO$_3^-$ (315 mg L$^{-1}$), and 0.0025 mol L$^{-1}$ SO$_4^{2-}$ (240 mg L$^{-1}$), which are higher than the anion concentration ranges typical of most freshwaters. 34, 40, 41

Table 2. Comparison of the DGT measurements for NH$_4$-N at different ionic strengths (mol L$^{-1}$ NaCl). Data are mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Ionic strength</th>
<th>$C_{\text{AMI-DGT}}$/$C_{\text{SOLN}}$</th>
<th>$C_{\text{A520E-DGT}}$/$C_{\text{SOLN}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.001</td>
<td>1.01 ± 0.13</td>
<td>1.09 ± 0.04</td>
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<tr>
<td>0.001</td>
<td>0.91 ± 0.04</td>
<td>1.06 ± 0.06</td>
</tr>
<tr>
<td>0.01</td>
<td>1.03 ± 0.05</td>
<td>0.95 ± 0.04</td>
</tr>
<tr>
<td>0.1</td>
<td>0.13 ± 0.004</td>
<td>0.08 ± 0.04</td>
</tr>
</tbody>
</table>

Figure 1. Comparison of the DGT measurements in solution with various concentrations of Cl$^-$ (A), HCO$_3^-$ (B) and SO$_4^{2-}$ (C) for 72 h deployments. $C_{\text{AMI-DGT}}$/$C_{\text{SOLN}}$ (grey) and $C_{\text{A520E-DGT}}$/$C_{\text{SOLN}}$ (black). Data are mean values (n = 3) ± 1 standard deviation. Experimental conditions: initial NO$_3$-N concentration = 0.90 ± 0.18 mg L$^{-1}$, final NO$_3$-N concentration = 0.84 ± 0.062 mg L$^{-1}$; pH = 7.0 ± 0.47 for 0.0005 - 0.012 mol L$^{-1}$ Cl$^-$, 0.0005 - 0.0005 mol L$^{-1}$ HCO$_3^-$ and 0.0005 - 0.0025 mol L$^{-1}$ SO$_4^{2-}$. pH = 8.4 ± 0.27 for 0.005 and 0.012 mol L$^{-1}$ HCO$_3^-$ and pH = 5.8 ± 0.29 for 0.005 mol L$^{-1}$ SO$_4^{2-}$.
3.5 Cationic interferences with NH$_4$-N measurements by CMI-DGT

The effects of Na$^+$, K$^+$, Ca$^{2+}$, and Mg$^{2+}$ on NH$_4$-N measurements by CMI-DGT were investigated and compared with PrCH-DGT by deploying DGT samplers of each type for 72 h in solutions with varying cation concentrations (Figure 2). $C_{\text{DGT}}:C_{\text{SOLN}}$ ratios for NH$_4$-N tended to decrease with increasing concentration of all cation species for both CMI-DGT and PrCH-DGT, with divalent Ca$^{2+}$ and Mg$^{2+}$ having greater impacts relative to that of Na$^+$ and K$^+$. $C_{\text{DGT}}:C_{\text{SOLN}}$ ratios were between 0.8 and 1.1 for CMI-DGT and PrCH-DGT at 0.0005 and 0.002 mol L$^{-1}$ Na$^+$ and K$^+$, 0.0003 and 0.0012 mol L$^{-1}$ Ca$^{2+}$, and 0.0003 and 0.0015 mol L$^{-1}$ Mg$^{2+}$. At cation concentrations above these levels both DGT methods grossly underestimated NH$_4$-N solution concentrations. Our results indicate that competing cations interfered with two binding layers differently. The affinity of CMI-DGT for competing cations decreased in the order Mg$^{2+} >$ Ca$^{2+} >$ K$^+ >$ Na$^+$ and PrCH-DGT for Ca$^{2+} >$ Mg$^{2+} >$ K$^+ >$ Na$^+$. Further investigations (data not shown) determined the concentration ranges for Ca$^{2+}$ for CMI-DGT were up to 0.0015 mol L$^{-1}$ (60 mg L$^{-1}$) and for PrCH-DGT were up to 0.0008 mol L$^{-1}$ (32 mg L$^{-1}$). Both DGT methods can accurately measure NH$_4$-N up to 0.002 mol L$^{-1}$ Na$^+$ (46 mg L$^{-1}$) and K$^+$ (78 mg L$^{-1}$), 0.0003 or 0.0012 mol L$^{-1}$ Ca$^{2+}$ (12 or 48 mg L$^{-1}$) and 0.0015 mol L$^{-1}$ Mg$^{2+}$ (36 mg L$^{-1}$). These levels are higher than general cation concentrations in most freshwaters, both methods are suitable for 72 h deployment.$^{34,40,41}$

![Comparison of the DGT measurements in solution with various concentrations of Na$^+$ (A), K$^+$ (B), Ca$^{2+}$ (C) and Mg$^{2+}$ (D) for 72 h deployment: $C_{\text{DGT}}:C_{\text{SOLN}}$ (grey) and $C_{\text{DGT}}:C_{\text{SOLN}}$ (black). Data are mean values (n = 3) ± 1 standard deviation. Experimental conditions: initial NH$_4$-N concentration = 0.6 ± 0.1 mg L$^{-1}$, final NH$_4$-N concentration = 0.44 ± 0.12 mg L$^{-1}$; pH = 7.0 ± 0.37 for 0.0005 - 0.03 mol L$^{-1}$ Na$^+$ and K$^+$, 0.0003 - 0.0025 mol L$^{-1}$ Ca$^{2+}$ and 0.0003 - 0.003 mol L$^{-1}$ Mg$^{2+}$; pH = 5.8 ± 0.36 for 0.005 mol L$^{-1}$Ca$^{2+}$ and 0.006 mol L$^{-1}$ Mg$^{2+}$.](image)

3.6 Intrinsic binding capacity of AMI-DGT and CMI-DGT

To accurately determine the concentration of an analyte in solution using DGT method, the mass accumulated during the measurement should not approach the binding capacity of the adsorbent. The binding capacity of the membrane-based DGTs for NO$_3$-N and NH$_4$-N were investigated by deploying nine sets of triplicate DGT samplers of each type in a 13.5 mg L$^{-1}$ NO$_3$-N and 30 mg L$^{-1}$ NH$_4$-N solution up to 72 h (Figure S3). The mass of NO$_3$-N and NH$_4$-N accumulated by the AMI and CMI membranes concurred with the predicted accumulated mass of NO$_3$-N and NH$_4$-N over the first 40 h and 56 h, respectively. After that, accumulation declined and appeared to approach their capacity values. The intrinsic binding capacities of AMI-DGT and CMI-DGT were approximately 921 ± 88 μg and 3512 ± 51 μg for NO$_3$-N and NH$_4$-N, respectively. The capacity of AMI-DGT was about 8.5% higher than A520E-DGT (849 ± 24 μg)$^{38}$ and the capacity of CMI-DGT were approximately 18% higher than PrCH-DGT (2990 ± 194 mg).$^{10}$ The results showed that the intrinsic binding capacities of membrane-based DGTs for NO$_3$-N and NH$_4$-N were higher than resin-based DGTs.

3.7 Evaluate performance in synthetic freshwater

DGT samplers of each type with a δh of 0.09 cm were deployed in synthetic freshwater with relatively low concentrations of NO$_3$-N (0.3 mg L$^{-1}$) and NH$_4$-N (0.1 mg L$^{-1}$) to evaluate their performances (Figure 3). The performance of AMI-DGT was compared with A520E-DGT and CMI-DGT was compared with PrCH-DGT. The results indicated there was a good agreement between the mass of NO$_3$-N by AMI-DGT and the theoretical mass estimated using the DGT equation over 72 h deployment in synthetic freshwater. The results of AMI-DGT and A520E-DGT over time in synthetic freshwater were also highly similar. The solution NH$_4$-N concentrations for the CMI-DGT and PrCH-DGT experiment changed over time and therefore the theoretical mass curves were not linear and the curve of CMI-DGT was more obvious due to the decreasing NH$_4$-N concentration in the solution. The mass of NH$_4$-N accumulated by CMI-DGT matched the theoretical mass over 48 h. After 48 h the mass of NH$_4$-N accumulated by CMI-DGT tended to be slightly lower than the theoretical mass. PrCH-DGT provided a better match between the mass of NH$_4$-N accumulated by DGT and the theoretical mass over 72 h deployment. These results differ based on other validation findings of CMI-DGT, which can be due to the competition of mixed ions in synthetic freshwater. Overall, the results demonstrated that membrane-based DGTs were able to quantitatively measure NO$_3$-N and NH$_4$-N concentrations under the physicochemical conditions typical of freshwater over 72 h deployment.

3.8 Field performance of DGT in freshwater

Measurements of NO$_3$-N and NH$_4$-N concentrations by AMI-DGT and CMI-DGT were evaluated at two field sites on the Gold Coast, Queensland, Australia. These measurements were compared to NO$_3$-N and NH$_4$-N determined by A520E-DGT and PrCH-DGT and to the mean grab sample concentrations obtained daily over the same deployment periods. The limits of detection (estimated as 3 × the standard deviation of the field blanks, n = 3) for AMI-DGT and A520E-DGT for NO$_3$-N with
a Δg of 0.09 cm were 2.02 µg L⁻¹ and 3.33 µg L⁻¹, and CMI-DGT and PrCH-DGT for NH₄-N with a Δg of 0.13 cm were 0.31 µg L⁻¹ and 0.42 µg L⁻¹, respectively, based on a 72 h deployment. Compared with resin gel-based DGTs, membrane-based DGTs had lower detection limits, likely due to their more consistent compositions and lower contamination.

These observations reinforce the limitations of monitoring nutrients using infrequent grab samples. The daily sampling appears to capture the changes in the nutrient concentrations quite well, although there is no way to be confident that the highest concentrations or even all of the variations have been captured. Although DGT measurements are made less frequently, the results follow the changing nutrient concentrations quite well, especially for NO₃-N (Figure 4). Most importantly, DGT responds to all the changes in nutrient concentrations, including those not apparent from the grab samples. The NH₄-N concentrations measured by DGT are sufficiently different from the grab samples for us to suggest that important data are missing from the grab sample record, especially during the prolonged rainfall event.

DGT measurements with the membrane and resin-based binding layers were not significantly different at Loders Creek (t-test, p > 0.05). Membrane-based DGTs and resin gel-based DGTs provided highly representative results for NO₃-N and NH₄-N concentrations. A closer examination confirmed that they closely followed the trends in concentrations measured in grab water samples over the nine days at Loders Creek (Tables S3 and S4). The C₅₂₀₋₄₀-DGT/C_SOLN and C₅₋₋₄₀-DGT/C_SOLN were between 1.28 and 0.98, and 1.26 and 0.88 during each DGT deployment, although from 8th to 11th December the C₅₋₋₄₀-DGT/C_SOLN and C₅₋₋₄₀-DGT/C_SOLN ratios were slightly high at 1.3 and 1.4, respectively. There was no significant difference between C₅₋₋₄₀-DGT and C₅₋₋₄₀-DGT measured, and C_SOLN concentrations measured in the grab samples over the DGT deployment period (t-test, p > 0.05). This may be due in part to the variation in grab sample concentrations over 72 h. Although the DGT measurements were not significantly different, there were interesting trends in the data. In all comparisons the resin-based DGT produced higher results. Although this was very minor for NO₃-N (Table S3), the results for NH₄-N had a lower ratio (Table S4) which indicated that PrCH resin-based DGTs are better than CMI membrane-based DGTs.
DGT samplers were also deployed at Saltwater Creek from 27th January to 5th February 2015. The conductivity in Saltwater Creek was ~0.36 mS cm⁻¹ from 27th – 30th January, increased to ~0.40 mS cm⁻¹ from 31st January to 2nd February and decreased to ~0.30 mS cm⁻¹ over the last three days of the study period. The NO₃-N concentration in the grab water samples from Saltwater Creek was 300 µg L⁻¹ on 27th January, decreased steadily to ~130 µg L⁻¹ on 31st January and then increased again to 210 µg L⁻¹ on 3rd February following a moderate rainfall event (Figure 5), then remained relatively stable for the rest of the sampling period. Similarly, NH₄-N concentration measured in grab samples also declined from ~29 µg L⁻¹ on 27th January to ~16 µg L⁻¹ between 27th January and 3rd February, then fluctuated around this value until the end of the sampling period. High NO₃-N and NH₄-N concentrations in grab samples were observed on 27th January, which could be due to a heavy rainfall event (156 mm) two days before the initial deployment (data sourced: Australian Bureau of Meteorology). After that, both NO₃⁻ and NH₄⁺ concentrations declined steadily over the dry period to 31st January. The moderate rainfall on the 1st February (18.2 mm) and 2nd February (14.2 mm) seemed to increase both NO₃⁻ and NH₄⁺ concentrations (with the latter peaking more quickly). NO₂⁻-N remained quite stable over the next few days while NH₃-N concentrations decreased quickly and were much more variable. Comparing the results from both sites, rainfall is obviously a major influence on dissolved inorganic nutrient concentrations with NO₃⁻-N tending to increase after rainfall, while NH₃-N was much less predictable. Nutrient concentrations in the Saltwater Creek were much higher than the Queensland Water Quality Guidelines value for NO₃⁻-N but varied around the guideline value for NH₃-N.

These results again indicated the limitations of monitoring nutrients using infrequent grab samples as the nutrient concentrations were different at each sampling time with rain or without rain. Even weekly or daily sampling would be insufficient to obtain a representative sample with concentrations varying by up to 400%. In comparison, the DGT measurements do not demonstrate the overall variability as well, but did respond to all changes in nutrient concentrations over nine-day deployment, including events or changes missed by the grab water samples. AMI-DGT and A520E-DGT provided similar (CAMI-DGT:CAS520E-DGT between 1.06 - 1.1) and highly
representative results for NO$_3$-N during the nine-day deployment (Table S5). Independent t-tests indicated that there was no significant difference among values of the DGTs and grab samples ($p > 0.05$) for NO$_3$-N. Although the results for NH$_4$-N from CMI-DGT and PrCH-DGT were more variable, independent t-tests confirmed that there was no significant difference between the two methods ($t_{CMI-DGT-PrCH-DGT}$ between 1.1 - 1.2) (Table S6) or CMI-DGT and PrCH-DGT and the average values of grab samples ($p > 0.05$).

4. Conclusions

New DGT techniques to measure NO$_3$-N and NH$_4$-N in freshwaters, using ion exchange membranes as binding layers, were successfully developed and validated. These binding layers exhibited similar, satisfactory characteristics as previously described resin-based binding layers, with the same functional groups and can be deployed for at least 72 h in waters with conductivity $\leq 1$ mS cm$^{-1}$. As observed previously, the NH$_4$-N diffusion coefficient through the agarose hydrogel changed with ionic strength (measured as conductivity), which necessitates the measurement of conductivity over the deployment period. Another study has used polyacrylamide with a lesser degree of variability with ionic strength and these binding layer types will be evaluated in future research.

The new DGT techniques performed well in field studies over nine days using back-to-back 72 h deployments in two local freshwaters systems. There was no significant difference between these DGT methods and the average concentrations determined from daily filtered grab samples and also with the previously described resin-based DGT techniques. Despite there being no significant differences between the DGT techniques with membrane and resin-based binding layers, there were clear tendencies, especially for NH$_4$-N, where one binding layer gave higher concentrations, although the binding layer that gave higher concentrations varied between the two freshwaters. This may in part be due to the slightly different curves used to calculate $D$ for each binding layer; the potential to use polyacrylamide diffusion layers may therefore see improved agreement in the future. The membrane and resin-based binding layers also experience slightly different competition effects from major ions, which may mean that a preference will be established in certain types of freshwaters. Use of commercial membranes as binding layers has several advantages. They are easier to prepare, avoid the use of toxic chemicals and have a consistent composition because they are produced commercially and have lower detection limits compared with resin-based DGT. AMI-DGT and CMI-DGT are excellent alternative methods to AS20E-DGT and PrCh-DGT to measure NO$_3$-N and NH$_4$-N, respectively, in most freshwaters. Future work will evaluate the application of AMI-DGT and CMI-DGT in measuring nutrient loads in freshwater systems.

Acknowledgements

We thank the School of Environment, Griffith University, for providing a Ph.D. scholarship and funding for J. H.

Supplementary information

Results: Comparison of the uptake and elution efficiencies between binding membranes and hydrogels; the uptake at different pH between binding membranes and hydrogels; relationship between the diffusion coefficient of NH$_4$-N and conductivity for CMI-DGT; binding capacity of AMI-DGT for NO$_3$-N and CMI-DGT for NH$_4$-N; NO$_3$-N and NH$_4$-N concentrations at Loders Creek and Saltwater Creek.

References

5. Diffusive gradients in thin films techniques provide representative time-weighted average measurements of inorganic nutrients in dynamic freshwater systems

This fourth results chapter addresses the third aim of the thesis to systematically evaluate the DGT techniques as a monitoring tool for inorganic nutrient concentrations in freshwater systems. The A520E-DGT for nitrate and PrCH-DGT for ammonium developed in chapters 2 and 3, and the existing Metsorb-DGT technique for phosphate were used to monitor the dissolved inorganic nutrient concentrations in various freshwater systems representing a range of settings, and hydrological and physicochemical conditions. The chapter therefore provides a true field validation of the utility of DGT techniques for routine monitoring of dissolved inorganic nutrients in freshwater systems.

This chapter has been submitted to *Environmental Science and Technology* and formatted in the journal style.

The co-authors of this manuscript are my thesis supervisors, Prof. Peter Teasdale, Assoc. Prof. David Welsh and Dr. William Bennett. They provided feedback on experimental design and manuscript drafts. PhD student Tianling Li provided help with field experiments. My (Jianyin Huang) contribution to the manuscript involved: devising the experimental design, all data collection and analysis, and preparation of the manuscript.
Jianyin Huang

Corresponding Author: Assoc. Prof. David Welsh

Principal Supervisor: Prof. Peter Teasdale

Principal Supervisor: Assoc. Prof. David Welsh
Abstract

Nutrient concentrations in freshwaters are highly variable over time, with changes driven by weather events, anthropogenic sources, modifications to catchment hydrology or habitats, and internal biogeochemical processes. Measuring infrequently collected grab samples is unlikely to represent nutrient concentrations in such dynamic systems. In contrast, in situ passive sampling techniques, like the diffusive gradients in thin films (DGT) technique, provide time-weighted average analyte concentrations over the entire deployment time. Two recently developed DGT techniques for nitrate (A520E-DGT) and ammonium (PrCH-DGT), as well as the Metsorb-DGT technique for phosphate, were used to monitor DGT-labile inorganic nutrients in various freshwater systems. Frequent grab sampling showed that concentrations of NH$_4^-$-N and NO$_3^-$-N changed dramatically in most of the studied freshwater systems over short timescales, while there were only relatively small fluctuations in PO$_4^{3-}$-P. The DGT measurements were highly representative when compared with the average nutrient concentrations obtained from daily grab samples over short-term (24 h) and long-term (72 h) deployments. DGT-labile concentrations were typically 100 - 112% of the average concentrations from grab samples collected over the deployment periods. The results of this study confirmed that DGT measurements provided a reliable and robust method for monitoring NH$_4$-N, NO$_3$-N and PO$_4$-P in a diverse range of dynamic freshwater systems.
1. Introduction

Nitrogen (NH$_4$-N and NO$_3$-N) and phosphorus (PO$_4$-P) are the primary nutrients that determine the productivity of aquatic ecosystems. However, due to human activities like the widespread use of fertiliser,$^{1,2}$ release of treated and untreated sewage$^3$ or industrial effluent,$^4$ and increased use of specific household products,$^5$ nutrient concentrations in aquatic environments have increased dramatically over the last few decades.$^6,7$ Excess nutrient loadings to rivers, lakes, bays and estuaries have led to eutrophication, the associated problems of phytoplankton and algal blooms, and subsequent episodes of water column hypoxia or anoxia when these blooms collapse and decompose.$^6,8$ The ecological health of natural waters, particularly freshwater, has become a global concern and many countries have developed guidelines to protect against the effects of eutrophication.$^9-12$ Liu et al. modelled the past and future dissolved inorganic nitrogen (DIN: NH$_4$-N, NO$_3$-N and NO$_2$-N) and phosphorus (DIP: PO$_4$-P) in the world’s major rivers. The results indicated that the number of rivers polluted by DIN and PO$_4$-P had increased by 33% and 25%, respectively, from 1970 to 2000 and predicted that DIN and PO$_4$-P would increase by a further 38% and 77%, respectively, by 2050. These circumstances clearly demonstrate the need to establish sophisticated monitoring programs for DIN and PO$_4$-P using accurate and representative measurements.

Nutrient loads in rivers and estuaries can vary significantly across a wide range of temporal and spatial scales. In order to assess the quality of water sources and/or the likelihood of eutrophication, nutrient loads (estimated as the product of concentrations and water flow)$^{13}$ have to be determined. However, dissolved inorganic nutrient concentrations can differ spatially due to natural differences in climate,$^{14}$ habitats, catchment geology and hydrology; anthropogenic factors such as habitat clearing and agriculture;$^{15}$ modifications to catchment
hydrology;\textsuperscript{16} and the presence of point sources such as wastewater treatment plants.\textsuperscript{17} Additionally, nutrient concentrations can also change considerably over time due to events such as stormwater runoff,\textsuperscript{18} effluent release, daily shifts in biological productivity from net photosynthesis to respiration,\textsuperscript{19, 20} and major N-cycling processes such as ammonification, nitrification and denitrification.\textsuperscript{21} This variability over time provides challenges to interpreting the results of monitoring as it may occur on a scale that obscures regional differences.\textsuperscript{22} Consequently, the widespread use of infrequent grab sampling methods for monitoring purposes is unlikely to be representative of nutrient concentrations within aquatic systems and may entirely miss high-concentration pulses.\textsuperscript{23, 24} Auto-sampler stations are now used to obtain and preserve more frequent grab samples, for instance in order to characterize changes in concentrations over a hydrograph,\textsuperscript{25} but these are very expensive and tend to be used at a small number of strategic locations. Therefore, there is a clear need for complementary and less expensive methods, such as passive samplers,\textsuperscript{26} to determine representative nutrient concentrations in freshwater systems.

\textit{In situ} passive sampling techniques provide a time-weighted average concentration over the deployment time, which varies from days to many weeks depending on the analyte.\textsuperscript{27} Because this time-integrated measurement responds to all concentration changes during deployment and integrates temporal environmental variability, it provides a more representative measurement of solute concentrations compared with grab sampling.\textsuperscript{28} Moreover, as analyte species are continuously accumulated within the passive sampling device, the concentration in the measured extract will usually be higher than from grab samples, reducing the uncertainty of analytical determination and improving detection limits.\textsuperscript{29} The diffusive gradients in thin films (DGT) technique is a well-established kinetic-regime passive sampling technique that relies on diffusion of analytes through a gel layer of
known thickness before accumulation on a layer containing an analyte specific binding agent. The DGT technique has previously been used to measure concentrations of various contaminants, including trace metal ions,\textsuperscript{30, 31} dissolved sulphide,\textsuperscript{32, 33} and oxyanions such as PO\textsubscript{4}-P, As, V and Sb.\textsuperscript{34-36} Several DGT techniques have been developed for the determination of PO\textsubscript{4}-P,\textsuperscript{37, 38} including Metsorb-DGT,\textsuperscript{39} which utilises a titanium dioxide-based binding agent. Recently, DGT techniques for the measurement of NO\textsubscript{3}-N (A520E-DGT)\textsuperscript{40} and NH\textsubscript{4}-N (PrCH-DGT)\textsuperscript{41} in freshwaters have been developed, which use ion exchange resins as binding agents. This study focuses on evaluating the capability of these three DGT techniques to measure representative dissolved inorganic nitrogen and phosphorus concentrations in seven dynamic freshwater systems on the Gold Coast, Queensland, Australia.

2. Experimental Section

2.1 Description of study locations

Seven freshwater sites within five urban waterways on the Gold Coast, Queensland, were used for this study (Figures S1 - S2). These included Saltwater Creek (stream and wetland), Loders Creek (stream), Gold Coast Regional Botanic Gardens (wetland), Currumbin Creek (stream and wetland) and Worongary Creek (stream).

Saltwater Creek is a micro-tidal stream with an urbanised and modified freshwater catchment.\textsuperscript{42} The stream is approximately 17 km long, 13.5\% of which flows through suburban areas. An artificial pond, constructed as a sediment retention basin during the clearing and building stage of an estate adjacent to Saltwater Creek, which receives local stormwater run-off, was also used in this study. The pond is used for flood storage for Saltwater Creek during the wet season. Loders Creek is a micro-tidal stream with a total catchment area of approximately 10 km\textsuperscript{2}, which is dominated by light industry and well-
established residential areas. The freshwater section of Loders Creek has been highly modified to manage stormwater but the northern tributary, selected for this study, still has a seemingly natural channel. Previous studies have reported very high concentrations of NO$_3$-N and NH$_4$-N at this site.$^{40, 41}$ The Botanic Gardens is bordered by a golf club and a large residential area, and contains a series of artificial ponds, dense with lilies and algae, which receive stormwater run-off from the surrounding areas and the gardens. Previous measurements at this site have found high NO$_3$-N and NH$_4$-N concentrations. Worongary Creek is the northern tributary in the Mudgeeraba Creek catchment and is surrounded by a golf club and urban and rural residential areas. Currumbin Creek is a micro-tidal stream, which is approximately 24 km long, flowing from Mount Cougal National Park to Currumbin Alley. The stream sampling site was located downstream of the National Park in peri-urban Currumbin Valley with mostly acreage properties and small-scale agriculture. The wetland in this system is a pond where the stream channel widens close to the uppermost part of the estuary and is surrounded by acreage properties.

2.2 Field study design and methods

During the study period, pH, temperature, conductivity and dissolved oxygen were recorded 1 - 2 times daily (8 - 10 am and 2 - 4 pm) at each site, at a water depth of approximately 30 cm, using a calibrated combination meter (YSI ProPlus Multiparameter). Rainfall conditions (every 2 h) for each deployment were obtained from the Australian Bureau of Meteorology (www.bom.gov.au).

DGT samplers for each analyte were deployed at the same water depth mentioned above in triplicate ($n = 3$) for 24 h at four of the seven field sites (Saltwater Creek wetland, Loders Creek, Botanic Gardens and Currumbin Creek stream) and for 72 h at all seven field sites.
DGT samplers of each type were also deployed in triplicate (n = 3) for periods of approximately three days over nine consecutive days at Currumbin Creek. DGT samplers with diffusive layer thickness (Δg) of 0.09 and 0.13 cm were selected for the calculation of DGT determined concentrations (C_{DGT}) of the dissolved inorganic nutrients over 24 h (short-term) and 72 h (long-term) deployments, respectively. A deployment apparatus was designed, consisting of a plastic plate with holes to house the samplers and a backing plate to hold them in place, to deploy up to 12 DGT samplers at a time (Figure S3). The apparatus was anchored to a plastic pipe inserted into sediment so that the windows of the DGT probes faced the direction of water flow. Upon removal, the DGT samplers were rinsed briefly with deionised water, placed in plastic bags with 1 - 2 mL of deionised water and stored at 4 °C until analysis. Samplers for NO_3-N or PO_4-P with varying diffusive gel layer thicknesses (Δg) of 0.05, 0.09 and 0.13 cm (all deployed in triplicate) were deployed to allow calculation of the diffusive boundary layer thickness (DBL) (Figure S5). Field blank DGTs for each analyte were done in triplicate (n = 3) by exposing the laboratory blanks in the field for 30 seconds. The field blanks were then placed in sealed plastic bags and stored at 4 °C until analysis. Limits of detection of PrCH-DGT, A520E-DGT and Metsorb-DGT for NH_4-N, NO_3-N and PO_4-P were 8.70 µg L^{-1}, 11.6 µg L^{-1} and 2.12 µg L^{-1}, respectively, based on a 24 h deployment and a diffusive layer thickness of 0.09 cm; and 5.02 µg L^{-1}, 5.59 µg L^{-1} and 0.71 µg L^{-1}, respectively, based on a 72 h deployment and a diffusive layer thickness of 0.13 cm. All the field results were blank corrected.

Grab water samples were collected in triplicate 1 - 2 times daily (8 - 10 am and 2 - 4 pm) at each site to determine inorganic nutrient concentrations over the deployment periods. 50 mL syringes were rinsed 2 - 3 times with the site water before collecting the grab samples. Water samples were immediately filtered through 0.45 µm pore-size membrane filters (PVDF,
Millipore), stored in 50 mL conical centrifuge tubes (polypropylene, Sigma-Aldrich Australia), and kept cool until being frozen upon return to the laboratory.

2.3 General experimental

Deionised water (Milli-Q Advantage A10, 18.2 MΩ cm$^{-1}$) was used to prepare all solutions, and rinse all containers and materials used in this study. All chemicals were analytical reagent grade or equivalent. Ion exchange resins PrCH and A520E, and Metsorb (titanium dioxide) powder were provided by the Purolite Company and Graver Technologies, respectively. Ultrapure agarose (Life Technologies) and acrylamide (Bio-Rad) were used to prepare agarose and polyacrylamide diffusive and binding layers. Containers used to collect samples and for the preparation and storage of solutions, and the glass plates used for preparing gels, and DGT components were acid-cleaned in 10% (v/v) HCl (AR grade, Merck) for at least 24 h and rinsed thoroughly with deionised water prior to use.

2.4 DGT preparation and measurements

2.4.1 Preparation of diffusive and binding gel layers

Polyacrylamide diffusive layers for NO$_3$-N and PO$_4$-P DGT samplers were prepared as described previously$^{39}$ and stored in 0.001 mol L$^{-1}$ NaCl$^{40}$ and 0.01 mol L$^{-1}$ NaNO$_3$, respectively, after washing 2 - 3 times in deionised water. Ultrapure agarose (Life Technologies) diffusive layers for NH$_4$-N were prepared as described previously and stored in 0.001 mol L$^{-1}$ NaCl solution.$^{41}$ PrCH, A520E, and Metsorb binding layers for NH$_4$-N, NO$_3$-N and PO$_4$-P respectively, were prepared as described previously.$^{35, 40, 41}$ All binding layers were washed 2 - 3 times and stored in deionised water prior to use.
2.4.2 DGT sampler assembly

0.45 µm polysulfone filter membranes (VWR Australia) were selected for PrCH-DGT and A520E-DGT, as polysulfone membranes do not contain any nitrogen, which could be a potential source of contamination. Membranes were soaked in deionised water for 24 h and washed in 5% (v/v) HCl for another 24 h, rinsed with deionised water and stored in 0.001 mol L\(^{-1}\) NaCl solution. 0.45 µm cellulose nitrate membranes (Whatman) were used for Metsorb-DGT samplers as described previously.\(^{39}\) They were prepared in the same manner but washed in 5% (v/v) HNO\(_3\) for 24 h, rinsed with deionised water and stored in 0.01 mol L\(^{-1}\) NaNO\(_3\) solution.\(^{39}\)

DGT sampler moldings (DGT Research Ltd.) were acid washed (10% HCl AR grade) and rinsed in deionised water (2 - 3 times), before assembly. DGT samplers were assembled as described previously.\(^{39-41}\) The probes were stored in sealed plastic bags with 1 - 2 mL of deionised water prior to deployment.

2.4.3 Sample analysis

PrCH and A520E binding gels were eluted in 2 mL of 2 mol L\(^{-1}\) NaCl for 24 h,\(^{40, 41}\) and Metsorb binding gels were eluted in 1.5 mL of 1 mol L\(^{-1}\) NaOH for 24 h.\(^{39}\) Eluents were diluted 5- to 10-times with deionised water to meet the acceptable concentration range of the analytical method. Prior to analysis, the pH of the Metsorb binding layer eluents were adjusted to a circumneutral pH using dilute HCl.

NH\(_4\)-N, NO\(_3\)-N and PO\(_4\)-P concentrations in eluents and filtered grab water samples were measured according to standard methods for the examination of water and wastewater from American Public Health Association (APHA) 4500-NH\(_3\) H, 4500-NO\(_3\) F, and 4500-P G using
a Seal AA3 segmented flow analyser. Calibration standards from 0 to 380 μg L\(^{-1}\) NH\(_4\)-N, NO\(_3\)-N and PO\(_4\)-P were prepared from 1000 mg L\(^{-1}\) NH\(_4\)Cl, NaNO\(_3\) and KH\(_2\)PO\(_4\) certified standard solutions (Merck). Ammonium sulfate ((NH\(_4\))\(_2\)SO\(_4\)) (AR, Merck), potassium nitrate (KNO\(_3\)) (AR, Chem Supply) and potassium phosphate (KH\(_2\)PO\(_4\)) (AR, Chem Supply) were used to prepare quality control standards (at 50 μg L\(^{-1}\)), which were analysed frequently throughout each analytical run. Recoveries of quality control standards were typically between 95 - 110%. The results from quality control standards were used to correct for any observed instrument drift. The instrument detection limits, calculated as three times the standard deviation of the blanks, were 2.3 μg L\(^{-1}\) for NH\(_4\)-N, 0.59 μg L\(^{-1}\) for NO\(_3\)-N, and 0.66 μg L\(^{-1}\) for PO\(_4\)-P, respectively (n = 10).

2.4.4 \(C\text{DGT}\) calculation and statistical comparison with grab samples

DGT-measured concentrations (\(C\text{DGT}\): ng mL\(^{-1}\); converted to μg L\(^{-1}\)) of nutrients were calculated using the DGT equation (Eq 1\(^{30}\)):

\[
C\text{DGT} = \frac{M \Delta g}{D A t}
\]  

Where, \(M\) is the mass of NH\(_4\)-N, NO\(_3\)-N or PO\(_4\)-P bound to the adsorbent (ng) corrected with the appropriate elution efficiency,\(^{39-41}\) \(\Delta g\) is the diffusive layer thickness, \(D\) is the diffusion coefficient of inorganic nutrient ion through the diffusive layer (cm\(^2\) s\(^{-1}\)), \(t\) is the deployment time (s) and \(A\) (3.14 cm\(^2\)) is the area of the probe exposed to solution. Values of \(D\) for NO\(_3\)-N, NH\(_4\)-N and PO\(_4\)-P were obtained from Huang \textit{et al.} and Panther \textit{et al.}, respectively.\(^{39-41}\) The value of \(D\) for NH\(_4\)-N was adjusted for the conductivity of the site water according to Huang \textit{et al.}\(^{41}\) Independent-t tests were used to compare the triplicate DGT measurements and average concentrations obtained from daily grab samples.
Additionally, the deployment of DGT samplers with different diffusive gel layer thicknesses allowed the thickness ($\delta$) of the diffusive boundary layer (DBL) to be calculated at each field site\textsuperscript{38}:

$$\frac{1}{M} = \frac{\Delta g}{DC_{DGT}At} + \frac{\delta}{DC_{DGT}At} \quad (2)$$

A plot of $1/M$ versus $\Delta g$ is a straight line with a slope ($m$) of $1/(DC_{DGT}At)$ and intercept ($b$) of $\delta/(DC_{DGT}At)$. Therefore, $\delta$ (Eq 3) and $C_{DGT}$ (Eq 4) can be calculated.

$$\delta = \frac{b}{m} \quad (3)$$

$$C_{DGT} = \left( \frac{1}{mAt} \right) \quad (4)$$

When the thickness of the DBL was included in the DGT calculations, a value of 3.8 cm\textsuperscript{2} was used for the sampling area, $A$, as described by Warnken et al.\textsuperscript{43}

### 3. Results and discussion

#### 3.1 Variability of physicochemical parameters

Physicochemical data at each site during the study period are presented in Figure 1, with the mean, standard deviation and relative standard deviation (RSD) of each parameter at each site given in Table S1. The average pH (Figure 1A) at the study sites ranged from 6.61 (Worongary Creek) to 7.47 (Saltwater Creek stream), indicating circumneutral pH conditions at all sites. The Saltwater Creek wetland was the most variable with a pH range of 6.53 to 7.47 and an RSD of 3.5%. Mean temperatures at the sites (Figure 1B) ranged between 14.4 °C (Currumbin Creek) and 19.3 °C (Saltwater Creek wetland). The greatest variations in temperature were observed at Saltwater and Currumbin Creek wetlands (6.7% and 6.8% RSD, respectively). Conductivity data is shown in Figure 1C. The two sites with the highest average conductivities (0.636 mS cm\textsuperscript{-1} at Loders Creek and 0.655 mS cm\textsuperscript{-1} at Saltwater Creek stream) also had the most variable conductivities (22.3% and 13.1% RSD, respectively). This
may be because these sites are impacted most by stormwater run-off due to the high proportion of impermeable surfaces in both catchments. Figure S4 shows the impacts of rainfall on the conductivity at Saltwater Creek and Saltwater Creek wetland. The conductivities at Saltwater Creek and Saltwater Creek wetland decreased during rainy days from 15th to 17th June (5 mm to 18 mm), and after the rain stopped the conductivities at both sites increased back to 0.613 and 0.488 mS cm\(^{-1}\), respectively. The two lowest average conductivities were at the two Currumbin Creek sites (0.109 and 0.145 mS cm\(^{-1}\)), which are clearly influenced by drainage from a large pristine national park and a lowland catchment with a much lower proportion of impermeable surfaces. Most importantly, all conductivities were less than 1 mS cm\(^{-1}\), which is the recommended maximum for use of the NO\(_3\)-N and NH\(_4\)-N DGT techniques.\(^{40,41}\)

Dissolved oxygen saturation (%) (Figure 1D) also varied considerably between sites, with mean values ranging from 49% (Saltwater Creek stream) to 103% (Currumbin Creek stream). All sites, except Currumbin Creek stream had an average DO below the Queensland Water Quality Guidelines lower value of 85% in lowland streams and 90% in freshwater lakes. Within-site variability was also high, with some sites exhibiting large relative changes (> 50%) in dissolved oxygen over the relatively short study period, with Loders Creek having an RSD of 24.2% and Currumbin Creek wetland of 18.4%. The generally under-saturated dissolved oxygen concentrations indicate that all the sites, with the exception of Currumbin Creek, were net heterotrophic. Whereas the short-term variability is likely related to the degree of biological productivity, changes in water flow (rexygenation) and/or elevated oxygen demands possibly linked to labile organic matter inputs.
The measurements of NO$_3$-N, NH$_4$-N and PO$_4$-P in frequently collected grab samples (n = 7 over ~3 days) from the seven study sites (Figures 2 and S6) were largely made for the purposes of characterizing the variability in the diverse waterways and for comparison with the DGT measurements of the same nutrients over the same periods.

The grab sample measurements demonstrated that PO$_4$-P was the least variable nutrient, but concentrations were observed to increase after sustained moderate rainfall at the two Saltwater Creek sites (Figures 2A and S6A). The concentrations of DIN species were both
more variable with rainfall, with NO₃-N increasing substantially after rainfall at the two Saltwater Creek sites. A similar increase was not observed at Worongary Creek (Figure 2C), however, likely because grab samples were not collected during the actual rainfall event and the catchment of Worongary Creek is less urbanised, which may limit both the volume of run-off and the NO₃-N load during rainfall events. This suggests that stormwater run-off is a major source of NO₃-N, at least in Saltwater Creek, as has already being confirmed in the estuarine section of this waterway.⁴² NH₄-N also responded to rainfall, but in a less consistent manner than NO₃-N, sometimes increasing immediately or after a lag period, and sometimes decreasing. Ammonification (mineralisation of organic nitrogen by heterotrophic bacteria) is a major source of NH₄-N within aquatic ecosystems.¹⁹, ²⁰, ⁴⁴ Rainfall events may have multiple effects, such as flushing NH₄-N accumulated in the water column during periods of low flow out of a system, resulting in lower concentrations; increasing the input of NH₄-N-rich run-off into a system, resulting in higher concentrations; and transporting labile organic matter into or out of a system, resulting in changes to NH₄-N regeneration rates.¹⁸, ⁴⁵ Therefore, it is not unexpected that rainfall events had differing influences on NH₄-N concentrations in the studied systems, given the diversity of waterbody types and catchment conditions investigated.

The very high concentrations of NH₄-N (＞1500 μg L⁻¹) and NO₃-N (＞400 μg L⁻¹) within Loders Creek (Figure 2B) were unable to be explained by run-off from a recent rainfall event. They could be the result of intensive organic nitrogen turnover and partial nitrification of the regenerated NH₄-N, or suggestive of an anthropogenic source, such as light-industry. The Loders Creek catchment is dominated by light-industry and NH₃ is commonly used as an industrial bleach or disinfectant.
Substantial daily differences were also observed for DIN concentrations at two of the wetland sites [Currumbin Creek wetland and the Botanic Gardens (Figures 2D and S6B)] during periods when rainfall was absent or very minor. At Currumbin Creek wetland (Figure 2D), the average values of NH$_4$-N, especially, and NO$_3$-N collected in the early morning (50 - 100 μg L$^{-1}$ and 35 - 50 μg L$^{-1}$, respectively) were much higher than those collected in the afternoon (10 - 30 μg L$^{-1}$ and 20 - 30 μg L$^{-1}$, respectively). Similar changes were also observed at the Botanic Gardens wetland, although NO$_3$-N was less dynamic over the latter measurements. These fluctuations in DIN concentrations in these wetlands are most likely to be due to diurnal shifts in the balance between autotrophic and heterotrophic processes in the wetlands.\textsuperscript{19, 21} During the night, heterotrophic respiration in the sediment would dominate and

![Figure 2. Mean values of NO$_3$-N (●), NH$_4$-N (■) and PO$_4$-P (▼) in grab samples at Saltwater Creek pond (A), Lodgers Creek (B), Worongary Creek (C) and Currumbin Creek wetland (D). Bars indicate rainfall.](image-url)
NH₄-N produced by ammonification would efflux to and accumulate in the water column. Additionally, partial nitrification of the effluxing NH₄-N during transport would lead to production and efflux of NO₃-N which would also accumulate in the overlying water.²⁰ Whereas, over the course of the day, photosynthesis by benthic microalgae, rooted macrophytes, their epiphytes and phytoplankton will occur and photo-assimilation of DIN (especially NH₄-N) into the primary producer biomass would both limit nutrient efflux from the sediment and consume the nutrients that had accumulated in the water overnight.¹⁹,²¹ The Saltwater Creek wetland seemed to be influenced more by rainfall associated run-off over the study period, but some changes in NH₄-N concentrations after the rainfall event towards the end of the monitoring period are also consistent with diel changes in internal biogeochemical cycling playing a role in regulating water column DIN concentrations.

### 3.3 Comparison of DGT-measurements with grab sample concentrations

Short-term (24 h, Δg = 0.09 cm) deployments were carried out at four sites (Saltwater Creek wetland, Loders Creek, Botanic Gardens and Currumbin Creek stream) and long-term (72 h, Δg = 0.13 cm) deployments at all seven sites. $C_{DGT}/C_{SOLN}$ ratios for each deployment are used to compare the concentrations obtained with each DGT method, with $C_{SOLN}$ calculated as the average of all grab sample concentrations determined over the DGT deployment period and $C_{DGT}$ as the average of the three replicate DGTS. Limits of detection of PrCH-DGT, A520E-DGT and Metsorb-DGT for NH₄-N, NO₃-N and PO₄-P measurements were 8.70 μg L⁻¹, 11.6 μg L⁻¹ and 2.12 μg L⁻¹, respectively, based on 24 h deployment with a 0.09 cm diffusive layer thickness, and 5.02 μg L⁻¹, 5.59 μg L⁻¹ and 0.71 μg L⁻¹, respectively, based on 72 h deployment with a 0.13 cm diffusive layer thickness. NH₄-N concentrations from 72 h deployments at Saltwater Creek were below the limit of detection. As the Saltwater Creek stream site had a high average conductivity of 0.655 mS cm⁻¹ the 72 h deployment time may
have resulted in competition from major cations that resulted in an underestimation, although
the average grab sample concentrations for this deployment (7.01 μg L\(^{-1}\)) are only marginally
higher than the LOD (5.01 μg L\(^{-1}\)). DBL corrections were made for DGT calculations at all
sites as outlined in Tables S4 - S6. All the field results were blank corrected.

\[ \frac{C_{DGT}}{C_{SOLN}} \]

ratios for NH\(_4\)-N, NO\(_3\)-N and PO\(_4\)-P were between 0.86 - 2.1, 0.87 - 1.4, and 0.64
- 1.2, respectively, across all sites (Figure 3), but most (21 out of 33) were within 0.8 - 1.2 (a
range selected to indicate acceptable agreement between methods). Independent t-tests were
used to analyse the DGT results and various grab sample measurements. This analysis
indicated that there were no significant difference (\(\alpha > 0.05\)) between the DGT results and
the average values from grab samples in most of the field sites, which demonstrates that the
DGT techniques provided accurate time-weighted average concentrations of dissolved
inorganic nutrients in these diverse systems.

However, exceptions were observed (\(p < 0.05\)) for four measurements, where the DGTs
determined higher concentrations of NH\(_4\)-N (\(\frac{C_{DGT}}{C_{SOLN}}\) between 1.6 and 2.2) and NO\(_3\)-N
(\(\frac{C_{DGT}}{C_{SOLN}}\) between 1.3 and 1.5) compared with average values of grab samples at the
Botanic Gardens and Currumbin Creek wetland sites. This is not unexpected as these two
sites experienced dramatic changes in grab sample concentrations at different times of day
and it would be expected that the night time concentrations of DIN would generally be higher
than those during the day (see discussion in section 3.2). Therefore, the DGT measurements
may have more effectively measured the average DIN concentrations than the grab samples,
which were collected only during the daytime. Consequently, the DGT measurements are
likely more representative of the true average concentrations than the averaged grab sample
concentrations. The other two exceptions were lower \(\frac{C_{DGT}}{C_{SOLN}}\) ratios for PO\(_4\)-P at Loders
Creek (0.86, p = 0.043) and Currumbin Creek (0.65, p < 0.01). While only minor variation was observed in the grab sample PO$_4$-P concentrations at both sites (RSD < 10%), this does not mean that lower concentrations did not occur at other times between grab sample collection.

![Graph A](image1.png)

![Graph B](image2.png)

![Graph C](image3.png)

**Figure 3.** The ratio of $C_{DGT}$ and $C_{SOLN}$ of NH$_4$-N (A), NO$_3$-N (B) and PO$_4$-P (C) at each field sites for 24 h (○) deployments at four field sites (SCP, LC, BG and CC) and 72 h (■) deployments at all field sites. SC: Saltwater Creek; SCP: Saltwater Creek pond; LC: Loders Creek; BG: Botanic Gardens; WC: Worongary Creek; CC: Currumbin Creek; CCW: Currumbin Creek wetland. SC did not contain the ratio of $C_{DGT}$ and $C_{SOLN}$ for NH$_4$-N because the field data was below the detection limit.

Although the measurements were not significantly different, high $C_{DGT}/C_{SOLN}$ ratios of NH$_4$-N were observed at Currumbin Creek (≈ 1.4 for both deployments) and NO$_3$-N at Saltwater Creek (≈ 1.4 for 72 h deployments). The DGT measurements of NO$_3$-N would have better captured the effect of the period of extended moderate rainfall that occurred during the
Saltwater Creek deployment (Figure S6A) than grab samples, as there were some large changes in NO$_3$-N concentrations between adjacent grab samples and it is unlikely that the grab sample times were coincident with the peaks or troughs in NO$_3$-N concentration over the rainfall event, or that the changes in concentration were linear between the sampling points. In contrast, the A520E-DGT technique is able to integrate shifting concentrations and provide time-weighted average concentrations that better represent the true average NO$_3$-N concentrations than the average obtained from intermittently collected grab samples. The NH$_4$-N concentrations measured in grab samples in Currumbin Creek also appear to be subject to diurnal variations (RSD > 25%) similar to those observed in the Currumbin Creek wetland, which would lead to the daytime collected grab samples underestimating the actual average concentration (see discussion in section 3.2). Therefore, each of these differences can be related to the lower representativeness of the grab sample measurements, compared to the time-weighted DGT measurements. Overall, the DGT techniques provided ‘recoveries’ of $112 \pm 20\%$, $105 \pm 17\%$ and $102 \pm 12\%$ of average values of grab samples for NH$_4$-N, NO$_3$-N and PO$_4$-P respectively, at those field sites and can therefore be considered to provide accurate measurements, but corrections for the DBL were necessary to obtain that level of accuracy.$^{39-41}$

This study has shown the high variability of nutrient concentrations over time in natural waters and the limitations of monitoring nutrients using infrequent grab samples. The grab samples only provide a snapshot of the nutrient concentrations at each sampling time, with no information provided between samples. Therefore, infrequent grab samples are less representative of nutrient concentrations within a waterway and likely to miss periodic increases in concentration due to a variety of events. DGT techniques are able to capture the changes associated with these events as they integrate all changes in nutrient concentrations.
over the deployment period. However, grab samples do provide valuable information on short-term variability within dynamic waterways provided they are frequent enough to capture this variability. In this regard the two methods are highly complementary. A study in which high frequency grab samples, collected every 3 h for 24 h for instance, compared with DGT measurements made over 24 h or over day and night periods would allow an even more detailed assessment of the accuracy and precision of these DGT techniques.

3.4 Time series deployment at Currumbin Creek

Back-to-back 72 h DGT deployments were completed at Currumbin Creek stream over nine consecutive days (Figure 4). DGT results were blank and DBL corrected. Grab samples for DIN and PO₄-P were also collected daily over these periods, including when the DGTs were deployed and collected, and rainfall data was obtained from BOM. In most instances the DGT measurements were intermediate between the grab sample concentrations over the same period. There were multiple rainfall events (3.1 - 24.9 mm) two days before the deployments commenced, which resulted in relatively high nutrient concentrations during the first deployment day. Nutrient concentrations were quite variable in Currumbin Creek. NH₄-N concentrations measured in grab samples were 30 µg L⁻¹ on 25th February and increased to 43 µg L⁻¹ on 26th February after a minor rainfall event (≈ 5 mm). Thereafter, NH₄-N concentrations declined and fluctuated around 20 µg L⁻¹ until the end of the sampling period, although they increased slightly (28 µg L⁻¹) during a heavy rainfall (45 mm) event. NO₃-N concentrations in the grab water samples decreased from 170 µg L⁻¹ on 25th - 26th February to about 100 µg L⁻¹ on 27th February, then increased to 150 µg L⁻¹ on 28th February with the heavy rainfall. NO₃-N concentrations then declined steadily from 100 µg L⁻¹ on 1st March to 53 µg L⁻¹ on 6th March. Changes in PO₄-P concentrations in grab samples were similar to NH₄-N, responding to rainfall in a similar manner, which had not been observed in the
previous fieldwork. \( \text{PO}_4^3- \text{P} \) concentrations were 10 µg \( \text{L}^{-1} \) on 25\(^{th} \) February and increased to 15 µg \( \text{L}^{-1} \) on 26\(^{th} \) February. Subsequently, \( \text{PO}_4^3- \text{P} \) concentrations declined to ≈ 6 µg \( \text{L}^{-1} \) before increasing with the heavy rainfall and fluctuating around 8 - 10 µg \( \text{L}^{-1} \) until the end of the sampling period.

![Figure 4](image). Changes in \( \text{NH}_4^+ - \text{N} \), \( \text{NO}_3^- - \text{N} \) and \( \text{PO}_4^3- - \text{P} \) concentration measured in grab samples (●) and DGT values (□) for \( \text{NH}_4^+ - \text{N} \) (A), \( \text{NO}_3^- - \text{N} \) (B) and \( \text{PO}_4^3- - \text{P} \) (C) at Currumbin Creek from 25 February to 6 March 2015. Data are mean values (n = 3) ± 1 standard deviation. The columns represent the rainfall (mm) during the deployment at Currumbin Creek. 25 - 28 February: pH = 6.88 ± 0.11; conductivity = 0.106 ± 0.01 mS cm\(^{-1} \); temperature = 22.9 ± 0.6 °C. 28 February - 3 March: pH = 6.86 ± 0.05; conductivity = 0.110 ± 0.02 mS cm\(^{-1} \); temperature = 23.7 ± 0.2 °C. 3 - 6 March: pH = 6.90 ± 0.04; conductivity = 0.109 ± 0.05 mS cm\(^{-1} \); temperature = 24.3 ± 0.6 °C.

Comparison of the mean concentrations showed that the DGTs provided similar concentrations compared with the average values of grab samples over each deployment period (Table S7). Independent t-tests showed that only two DGT results (\( \text{NH}_4^+ - \text{N} \) from 3\(^{rd} \) to
6th March and PO₄-P from 28th February to 3rd March) were significantly different to the average grab sample (p < 0.05) concentrations for the same periods; although the DGT results were still quite similar, the grab samples had a low standard deviation for these deployments. These differences are also apparent in Figure 4. Based on the comparisons in section 3.4 and previous studies, in which DGTs have been demonstrated to provide highly representative measurements, it is probable that there were changes in nutrient concentrations that were not accurately captured by the daily grab samples. For instance, the heavy rainfall event on 28th February could readily have produced a higher peak PO₄-P concentration than was measured with the grab samples. Additionally, given the diurnal changes in NH₄-N observed, particularly at the wetland sites (Figures 2C and 6B), lower or higher NH₄-N concentrations may have been present at different times of the day to when these grab samples were collected, especially as all the grab samples were collected in the morning. Other circumstances can also readily be foreseen, especially over longer deployment times, where the DGT measurements would be quite different to average concentrations in daily grab samples, as it is highly unlikely that the grab sampling times would coincide with the maxima or minima in the nutrient concentrations or those changes in nutrient concentrations would be linear. These observations reinforce the limitations of monitoring nutrients using infrequent grab samples, as is done routinely with monthly environmental monitoring. Only regular samples collected using auto-samplers would be able to provide data with a similar degree of accuracy and representativeness as the DGT techniques utilised in this study.

4. Conclusion

DGT techniques for NH₄-N, NO₃-N and PO₄-N were evaluated at seven freshwater sites (stream or wetland) with a range of water quality conditions (pH, temperature, conductivity and dissolved oxygen) and catchment features, and at one site over a time series deployment.
Quite different nutrient concentrations and processes were observed at each of the sites. As was expected, \( \text{PO}_4\text{-P} \) was the least variable nutrient although rainfall events often led to an increase in concentration. \( \text{NH}_4\text{-N} \) concentrations were affected unpredictably by rainfall, but strong diurnal cycling was apparent in productive wetland sites. \( \text{NO}_3\text{-N} \) almost always increased with rainfall events and diurnal cycling was also observed at wetland sites. Despite these varying nutrient concentrations, on most occasions there were no significant differences between mean DGT measurements and the average nutrient concentrations obtained from grab samples over the same period. Where significant differences were observed they were usually able to be explained by the grab sampling frequencies being insufficient to obtain a representative average result, such as at wetland sites with strong diurnal variations, where grab samples were only collected during the day or where the peak concentrations associated with rainfall events were likely missed.

This study has clearly demonstrated that the *in situ* DGT techniques provide time weighted-average concentrations that are highly representative of the inorganic nutrient concentrations in dynamic waters over both 24 h and 72 h deployment times. Only one DGT replicate measurement (for \( \text{NH}_4\text{-N} \)) was below the detection limit, which in any case was close to the average grab sample value. These DGT techniques are therefore very suitable to being used for monitoring purposes to determine accurate and representative nutrient concentrations and loads in a wide range of freshwater systems. More importantly, DGT techniques will be able to provide a powerful tool to monitor and understand the dynamic changes in nutrient concentrations that can occur in freshwater ecosystems.
Acknowledgments

The authors are grateful for the School of Environment, Griffith University, for providing a Ph.D. scholarship and funding for J. H. We also thank the Purolite Company (www.purolite.com) for the provision of the Purolite A520E resin and the Microlite PrCH resin in this study.

Supplementary information

Experiments: Preparation of Metsorb binding layers; Maps of the field sites; The deployment apparatus for DGT samplers; Variability of physicochemical parameters; Diffusion boundary layer (DBL) calculations and corrections; Grab sample measurements; Time series deployment at Currumbin Creek. Results: The mean, standard deviation (SD) and relative standard deviation (RSD) of pH, conductivity, temperature and dissolved oxygen (DO) concentrations (% saturation) at each site; the influence of rainfall events on conductivities at Saltwater Creek and Saltwater Creek pond; the percentage of dissolved oxygen in the early morning and in the afternoon at each field site; the calculation of DBLs; grab samples of DIN and PO₄-P at Saltwater Creek, Botanic Gardens and Currumbin Creek, and the rainfall events; DGT results of nutrient concentrations with and without DBL; nutrient concentrations of grab samples and DGT results at Currumbin Creek.

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6. Conclusions and future research opportunities

The major aims of this research project were to:

1) Develop new diffusive gradients in thin films (DGT) techniques for nitrate (NO$_3$-N) and ammonium (NH$_4$-N).

2) Compare the performance of these new DGT techniques with concentrations from analysis of grab sample collected regularly over the deployment period from freshwater systems.

3) Evaluate the new DGT techniques under diverse field conditions, alongside the previously described phosphate (PO$_4$-P) DGT technique.

Aim 1 is addressed in Chapters 2, 3 and 4. Aim 2 is addressed in Chapters 2, 3, 4 and 5, and Aim 3 is addressed primarily in Chapter 5. The major findings are summarised below.

Chapters 2 and 3 describe the development and evaluation of new diffusive gradients in thin films techniques for nitrate and ammonium employing A520E (anion) and PrCH (cation) ion exchange resins, respectively, as the binding agents incorporated within polyacrylamide and agarose hydrogels, respectively. These were both selected from a short list after preliminary tests of several potential binding agents (zeolite, carbon nanotubes, layered double hydroxide Ni-Fe, Al$_2$O$_3$ and Powdex PCM resin). Each binding layer had high uptake and elution efficiencies, with the optimal eluent for both being 2 mol L$^{-1}$ NaCl. The A520E-DGT and PrCH-DGT quantitatively accumulated nitrate and ammonium over time with excellent
linearity, and A520E and PrCH binding layers had very high intrinsic binding capacities (without competing ions) for nitrate and ammonium, respectively. Both DGT techniques exhibited quantitative uptake over the pH range of 3.5 to 8.5, which is typical of the vast majority of freshwaters. Both DGT techniques were adversely affected by high ionic strengths (≈ 10 mmol L\(^{-1}\) and higher), which makes them inappropriate for use in estuarine or marine waters. The maximum concentrations of competing majors ions at which quantitative uptake was obtained were accurately determined and found to be higher than the average concentrations of global freshwaters, although PrCH-DGT measurements of NH\(_4\)-N in hard waters should be evaluated further. Using electrical conductivity as a proxy for major ion concentrations, both DGT techniques should be quantitative in freshwaters with conductivity < 1000 µS cm\(^{-1}\). Solution ionic strength (measured as conductivity) had a non-linear effect on the diffusion coefficient of NH\(_4\)-N in the agarose hydrogel diffusive layer, with decreasing conductivity leading to an increase in \(D\). A non-linear line was fit to diffusion coefficient (\(D\)) vs. conductivity data which was then used to obtain a \(D\) for solutions of particular conductivity. The PrCH-DGT technique was deployed in synthetic freshwaters for several days which indicated that the deployments should not be done for more than 72 h. Field deployments indicated the importance of determining the diffusion boundary layer (DBL) thickness and using that in the DGT calculations. It is recommended that DGTs with different diffusive layers are routinely deployed, so that the DBL can be calculated and taken into account when determining nutrient concentrations. Field studies confirmed that A520E-DGT and PrCH-DGT provided accurate and highly representative measurements of nitrate and ammonium concentrations during short-term deployments in freshwaters. For ammonium, the use of thicker diffusive layers was recommended for deployments longer than 24 h.
Chapter 4 described the development and evaluation of ion exchange membranes as alternative DGT binding layers for measuring nitrate (AMI) and ammonium (CMI) in freshwaters. Both membranes had high uptake and elution efficiencies similar to those for resin-based binding layers. The AMI and CMI membranes had high intrinsic binding capacities for nitrate and ammonium, respectively. The membrane-based DGTs are suitable to use in waters of pH 3.5 - 8.5 and over ionic strength ranges similar to the resin-based binding layers, but with slightly different upper limits for certain competing major ions. This suggests that the two DGT types may be better suited to measurements in freshwaters with a particular composition. The field experiments demonstrated that AMI-DGT and CMI-DGT can be an alternative to A520E-DGT and PrCH-DGT for measuring nitrate and ammonium, respectively, as they provided highly similar and representative results during the deployment time. Otherwise the membrane-based binding layers are easier to prepare and have a more consistent composition, which will make them preferred for some potential future applications.

Chapter 5 utilised A520E-DGT for nitrate, PrCH-DGT for ammonium and the existing Metsorb-DGT technique for phosphate, to determine dissolved inorganic nutrient concentrations in diverse freshwater systems. A520E-DGT and PrCH-DGT were selected based on their previous laboratory and field performances. A520E-DGT had very similar performances compared with AMI-DGT, however, laboratory and field results indicated that CMI-DGT underestimated the ammonium concentrations in synthetic freshwater and Loders Creek with high ionic strength, which differed from the other laboratory validations. The CMI-DGT performance needs to be further investigated. Nutrient concentrations at several field sites were observed to vary considerably over short time periods due to several different processes including stormwater run-off, anthropogenic contamination and diurnal shifts in
biogeochemical cycling, particularly for nitrate and ammonium. Even with these varying nutrient concentrations on most occasions there were no significant differences between mean DGT measurements and the average concentrations obtained from grab samples collected over the same period. Where significant differences were observed they were usually able to be explained by insufficient grab sample frequencies to obtain a representative average result. The results of this study confirmed that the new and the existing DGT techniques could provided accurate and highly representative methods for the routine in situ monitoring of ammonium, nitrate and phosphate in dynamic freshwater systems.
A number of areas for future research were identified from this thesis:

1. The contribution to nutrient loads of point and non-point sources is a major ecological concern and represents one of the most significant current and future water quality issues. The DGT techniques developed in this study can be used as a monitoring tool and are highly suitable for determination of nutrient loads in freshwater streams and rivers, including those that drain into estuaries. Nutrient loads are estimated as the product of the individual nutrient concentrations and flow rates, and are therefore strongly influenced by the accuracy and representativeness of the nutrient concentration data. Time-weighted average nutrient concentrations measured using the developed DGT techniques would provide much more representative data for load determinations than intermittent grab water samples and therefore improve load estimates. Such information would be invaluable to managers both to assess the quality of water resources and the likelihood of eutrophication in downstream ecosystems.

2. There is significant potential to utilise the DGT techniques developed in this study to determine bioavailable nutrient concentrations in soils, building on the previous research on soil phosphate. Nitrogen is a primary nutrient for plant growth and excess fertiliser use in agricultural settings is the major cause of nutrient pollution in waterways. Therefore, there are potentially significant economic and environmental benefits in utilising the developed DGT methods to monitor the nutrient status of agricultural soils. This may allow farmers to adjust the timing and quantity of fertiliser use to suit the growth needs of their crops and this in turn would reduce nutrient loads from agricultural practises to adjacent waterways.
3. The developed DGT methods could be used to measure nutrient profiles and two-dimensional distributions as part of studies of sediment biogeochemistry. Most significantly these methods can be used alongside other DGT or related DET (diffusive equilibration in a thin film) sediment techniques to determine the influences of heterogeneity on such processes, which is a major gap in current measurement capabilities.

4. There is an urgent need to develop DGT or other passive sampling techniques that can be utilised to monitor nitrate and ammonium in estuarine and marine environments. Anthropogenic nutrient loads from urban and agricultural areas, eventually end up in the marine environment, where they fuel eutrophication of estuarine and coastal waters, leading to increased primary production (so called green tides), degraded water quality and poor ecosystem health. Although, the A520E-DGT and PrCH-DGT, and ion exchange membrane-DGT techniques were developed during this study performed well in freshwaters, they were all negatively affected by high solution ionic strength, which makes them unsuitable for estuarine and marine waters. If new DGT methods can be developed and applied in saline waters, these would have significant implications for the monitoring of nutrients in marine systems.
Appendices

Appendix 1. Thesis as a series of published and unpublished papers

HDR Candidates may organise their thesis as a series of papers. This thesis format may include one or more papers that have been prepared, submitted, or accepted for publication. A Doctoral or MPhil thesis prepared in this way is not a different degree. There are several advantages to organising a thesis in this way:

- Preparing papers for publication saves time when preparing the thesis for examination as papers may make up one, or several, chapters within the thesis.
- It is to your advantage to publish work from your thesis as a means of disseminating your research, and developing your writing skills.
- It may improve the quality of your thesis as part of your thesis has already been subjected to peer review.
- Examiners may have more confidence in your thesis if they can see that you have already published your research. In addition, you will have already met one of the criteria of examination, with the thesis suitable for publication.

In addition, as a candidature requirement all doctoral candidates are expected to have at least one peer reviewed output accepted for publication during candidature. Whilst not compulsory, students are encouraged to include this publication in the body of the thesis due to the advantages as outlined above.

Conditions

Requirements for inclusion of papers

A thesis may include published or unpublished papers where such papers have been produced under supervision and during the period of candidature, and where the quality of such papers is appropriate to Doctoral, or MPhil, level research. For the purpose of this requirement, papers are defined as a journal article, conference publication, book or book chapter. Papers which have been rejected by a publisher must not be included unless they have been substantially rewritten to address the reviewers' comments, or have since been accepted for publication.

Overall, the material presented for examination needs to equate to that which would otherwise be presented in the traditional thesis format. This remains a matter of professional judgment for the supervisor and the student.

Extent of student’s contribution

The student should normally be principal author (that is, responsible for the intellectual content and the majority of writing of the text) of any work included in the body of the thesis. Where any work has been jointly authored, a signature from the corresponding author is required in order to include the material in the body of the thesis. Co-authored work in which the student was a minor author can only be used and referenced in the way common to any other research publication cited in the thesis.
Copyright

If copyright on a publication has been, or will be, assigned to a publisher, permission must be sought to reproduce the work in the thesis and allow for a digital copy to be made available on the institutional repository. This can be requested via the publication agreement with the publisher. Information on how to seek permission is available at:

» Your published articles

Most publishers will allow authors to store post-prints (submitted versions of papers to publishers with no publisher format) in institutional repositories. To enable this students should add an addendum to the publication agreement with publishers as follows (even if it is handwritten onto the agreement):

I seek your permission to archive a post-print of this publication in a public access institutional repository after peer review. By agreeing to publish my paper you consent to this addendum.

Further advice on how to seek permission from a publisher is detailed in the Copyright Guide.

If permission cannot be obtained, students may still include the publication in the body of the thesis, however following examination the relevant chapter(s) will be redacted from the digital copy to be held by the Griffith University Library so that the copyright material is not made publically available on the institution repository. Alternatively, students may ask for an embargo to be placed on the thesis in order to restrict public access to the thesis once the examination is complete.

Students are required to advise the copyright status of each publication included in the thesis via a declaration to be inserted in the thesis, as detailed below.

Students who require further advice regarding copyright issues are advised to contact INS Information Management.

General Enquiries Phone Number: (07) 3735 4007
(07) 3735 4007
General Enquiries Email: copyright@griffith.edu.au
Format

General
Consult the thesis preparation and formatting guidelines for general information about the requirements for formatting the thesis.

Format of papers
A thesis may entirely or partly be composed of papers. Where a thesis is entirely composed of papers, there is no minimum requirement for the number of papers that must be included and is a matter of professional judgment for the supervisor and the student. The papers may be rewritten for the thesis (post-print or pre-print versions): or they can be inserted in their published format, subject to copyright approval as detailed above. A paper may form a single chapter, or several papers may form successive chapters. Passages from papers may be transferred directly or in modified form into one or more of the chapters of the thesis. Students may repaginate the papers to be consistent with the thesis. However, this is at the discretion of the student.

Linking chapters
The thesis must be more than a collection of papers in the following ways. The chapters must be in logical order and strongly linked together. Students who submit a thesis in this format may introduce each new chapter with a foreword which introduces the research and establishes its links to previous chapters. In general, the thesis should include a general introduction which sets out the context of the thesis. The thesis should also include a conclusion which draws together the main findings of the thesis and establishes the significance of the work.

Declarations
All theses that include papers must include declarations which specify the publication status of the paper(s), your contribution to the paper(s), and the copyright status of the paper(s). The declarations must be signed by the corresponding author (where applicable). If you are the sole author, this still needs to be specified. The declaration will need to be inserted at the beginning of the thesis, and for any co-authored papers, additional declarations will need to be inserted at the beginning of each relevant chapter. You may wish to consult the declaration requirements for inclusion of papers diagram to ensure that you insert the correct declaration(s) within the thesis. Please note that completion of the declaration(s) does not negate the need to comply with any other University requirement relating to co-authored works as outlined in the Griffith University Code for the Responsible Conduct of Research.

Element requirements
Requirements for submitting a thesis as a series of published or unpublished papers may vary in some Elements. Consult your HDR Convener for further information.
Appendix 2. Supporting Information for the research article included as Chapter 2

Development and evaluation of a diffusive gradients in thin films technique for measuring nitrate in freshwaters

Jianyin Huang, William W. Bennett, Peter R. Teasdale*, Sean Gardiner, and David T. Welsh

* Environmental Futures Research Institute, Griffith University, Gold Coast campus, QLD 4215, Australia

* Corresponding Author: p.teasdale@griffith.edu.au. Tel: +61 (07) 555 28358. Fax: +61 (07) 555 28067
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Figure S1. Mass of NO$_3$-N accumulated by A520E-DGT over 24 h.

Figure S2. $C_{\text{DGT}}/C_{\text{SOLN}}$ for NO$_3$-N at various anion concentrations for 72 h deployment.

Figure S3. Determination of the capacity of A520E-DGT for NO$_3$-N.

Figure S4. Mass of NO$_3$-N accumulated by A520E-DGT in synthetic freshwater over 5 days.

Figure S5. Plot of reciprocal mass of NO$_3$-N accumulated by A520E-DGT versus diffusive layer thickness ($\Delta g$; cm) at Loders Creek (■) for the day 1 deployment and Saltwater Creek (○) for the day 1 - 2 deployment.

Table S1. The recipe of synthetic freshwater (pH = 7.20 ± 0.05).

Table S2. DBL thicknesses, and mean grab sample, and $C_{\text{DGT}}$ NO$_3$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Loders Creek.

Table S3. DBL thicknesses, and mean grab sample, and $C_{\text{DGT}}$ NO$_3$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Saltwater Creek.
Table S1. The recipe of synthetic freshwater (pH = 7.20 ± 0.05).

<table>
<thead>
<tr>
<th>Chemicals</th>
<th>Concentration (mmol L⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CaCl₂·2H₂O</td>
<td>0.375</td>
</tr>
<tr>
<td>Na₂SO₄</td>
<td>0.115</td>
</tr>
<tr>
<td>KCl</td>
<td>0.058</td>
</tr>
<tr>
<td>NaCl</td>
<td>0.229</td>
</tr>
<tr>
<td>MgCl₂·6H₂O</td>
<td>0.294</td>
</tr>
<tr>
<td>HCO₃⁻</td>
<td>0.983</td>
</tr>
</tbody>
</table>

The high R² value (≥ 0.9967) of NO₃-N accumulation confirmed that the new DGT method met the assumption of the DGT equation of linear mass accumulation for 24 h.

Figure S1. Mass of NO₃-N accumulated by A520E-DGT over 24 h. Data are mean values (n = 3) ± 1 standard deviation. The solid line is a regression line of best fit (y = 19.89x + 8.3876, R² = 0.9967). Experimental conditions: initial NO₃-N concentration = 11.3 mg L⁻¹, final NO₃-N concentration = 10.9 mg L⁻¹; matrix = 0.002 mol L⁻¹ NaCl; conductivity = 0.467 ± 0.031 mS cm⁻¹; temperature = 24.3 ± 0.3 °C.
**Figure S2.** \( \frac{C_{\text{DGT}}}{C_{\text{SOLN}}} \) for NO\(_3\)-N at various anion concentrations for 72 h deployment, SO\(_4^{2-}\) (dark grey), H\(_2\)PO\(_4^-\) (white), HCO\(_3^-\) (light grey) and Cl\(^-\) (black). Data are mean values (n = 3) ± 1 standard deviation. Experimental conditions: initial NO\(_3\)-N concentration = 0.91 ± 0.12 mg L\(^{-1}\), final NO\(_3\)-N concentration = 0.84 ± 0.62 mg L\(^{-1}\); time = 72 h; pH = 7.0 ± 0.47 for 0.0005 mol L\(^{-1}\) SO\(_4^{2-}\), H\(_2\)PO\(_4^-\), HCO\(_3^-\) and 0.0005 - 0.012 mol L\(^{-1}\) Cl\(^-\), pH = 5.8 ± 0.29 for 0.005 - 0.012 mol L\(^{-1}\) SO\(_4^{2-}\), pH = 5.1 ± 0.32 for 0.005 - 0.012 mol L\(^{-1}\) H\(_2\)PO\(_4^-\) and pH = 8.4 ± 0.27 for 0.005 - 0.012 mol L\(^{-1}\) HCO\(_3^-\).

Competition from the different anions became more serious over the longer deployment time.
Figure S3. Determination of the capacity of A520E-DGT for NO$_3$-N. Data points are mean values (n = 3) ± 1 standard deviation. The dashed line represents the predicted mass of NO$_3$-N accumulated by DGT. Experimental conditions: initial NO$_3$-N concentration = 13.6 mg L$^{-1}$, final NO$_3$-N concentration = 12.3 µg L$^{-1}$; conductivity = 0.36 ± 0.028 mS cm$^{-1}$.

The average mass of NO$_3$-N accumulated by the A520E binding gels deployed at 72 h was 849 ± 24 µg.
Figure S4. Mass of NO$_3$-N accumulated by A520E-DGT in synthetic freshwater over 5 days. Data points are mean values (n = 3) ± 1 standard deviation. The dashed line represents the predicted mass of NO$_3$-N accumulated by DGT. Experimental conditions: initial NO$_3$-N = 0.86 mg L$^{-1}$, final NO$_3$-N = 0.71 mg L$^{-1}$; conductivity = 0.277 ± 0.027 mS cm$^{-1}$; ion concentrations = 0.380 mmol L$^{-1}$ Ca$^{2+}$, 0.115 mmol L$^{-1}$ SO$_4^{2-}$, 0.058 mmol L$^{-1}$ K$^+$, 0.457 mmol L$^{-1}$ Na$^+$, 1.35 mmol L$^{-1}$ Cl$^-$, 0.150 mmol L$^{-1}$ Mg$^{2+}$ and 0.983 mmol L$^{-1}$ HCO$_3^-$; pH = 7.26 ± 0.07.

Figure S4 showed that the A520E-DGT was able to quantitatively measure solution NO$_3$-N concentrations over five deployment days.

Figure S5 showed example plots of 1/mass (ng$^{-1}$) versus Δg (cm) for A520E-DGT probes at each site. Good linearity was observed at each site (R$^2$ = 0.9652 at Loders Creek and R$^2$ = 0.9988 at Saltwater Creek for the examples shown in Figure S2) and the relative standard deviations were < 10%. Compared with Loders Creek, the changes of DBL for Saltwater Creek were more significant, especially for the day 4 - 6 deployment. The DBL ranged from 0.061 to 0.105 cm at Loders Creek and 0.038 to 0.141 cm at Saltwater Creek, which are
approximately 42 - 157% of the standard DGT diffusive layer thickness ($\Delta g = 0.09$ cm). $C_{\text{DGT}}$ values calculated taking the DBL into consideration excellent matches for the average concentrations measured in grab water samples collected over the deployment periods (Tables S3 and S4). However, when the DBL was not taken into consideration, $C_{\text{DGT}}$ values were gross underestimates of the average concentrations of NO$_3$-N measured in the grab samples. Therefore, it is suggested that DBL thicknesses are estimated for field deployments of the NO$_3$-N and other DGTs.

**Figure S5.** Plot of reciprocal mass of NO$_3$-N accumulated by A520E-DGT versus diffusive layer thickness ($\Delta g$; cm) at Loders Creek (■) for the day 1 deployment and Saltwater Creek (○) for the day 1 - 2 deployment of DGTs with varying diffuse gel thicknesses. Data points are mean values (n = 3) ± 1 standard deviation.

The DBL thickness changed over time at Loders Creek and Saltwater Creek. Compared with Loders Creek, the changes of DBL for Saltwater Creek was more significant, especially for the day 4 - 6 deployment, due to a rainfall event. The DBL ranged from 0.061 to 0.105 cm at Loders Creek and 0.038 to 0.141 cm at Saltwater Creek, which are approximately 42 - 157%
of the standard DGT diffusive layer thickness \( (\Delta g = 0.09 \text{ cm}) \). \( C_{DGT} \) values calculated with the DBL included provided excellent matches for the average values of grab samples collected over the deployments (Tables S3 and S4). However, there were large differences between \( C_{DGT} \) values and average concentrations of NO\(_3\)-N in the grab samples when the DBL was not taken into consideration. \( C_{DGT} \) values were underestimates when the DBL was not taken into consideration. Therefore, it is suggested that DBL thicknesses are estimated for field deployments of the NO\(_3\)-N and other DGTs.

### Table S2. DBL thicknesses, and mean grab sample, and \( C_{DGT} \) NO\(_3\)-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Loders Creek.

Data for NO\(_3\)-N concentrations are presented as mean values \((n = 3) \pm 1\) standard deviation.

<table>
<thead>
<tr>
<th>Day</th>
<th>DBL (cm)</th>
<th>( R^2 )</th>
<th>Average grab sample (( \mu g \text{ L}^{-1} ))</th>
<th>( C_{DGT} ) with DBL (( \mu g \text{ L}^{-1} ))</th>
<th>( C_{DGT} ) without DBL (( \mu g \text{ L}^{-1} ))</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.089 ± 0.039</td>
<td>0.9652</td>
<td>79.0 ± 31.2</td>
<td>80.1 ± 2.0</td>
<td>40.3 ± 1.0</td>
</tr>
<tr>
<td>2</td>
<td>0.105 ± 0.046</td>
<td>0.9576</td>
<td>56.3 ± 0.8</td>
<td>57.1 ± 3.0</td>
<td>26.4 ± 1.4</td>
</tr>
<tr>
<td>3</td>
<td>0.097 ± 0.041</td>
<td>0.9647</td>
<td>50.7 ± 7.1</td>
<td>52.3 ± 1.6</td>
<td>25.2 ± 0.8</td>
</tr>
<tr>
<td>4</td>
<td>0.061 ± 0.048</td>
<td>0.9371</td>
<td>31.9 ± 19.5</td>
<td>33.9 ± 4.6</td>
<td>20.2 ± 2.8</td>
</tr>
</tbody>
</table>
Table S3. DBL thicknesses, and mean grab sample, and $C_{\text{DGT}}$ NO$_3$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Saltwater Creek. Data for NO$_3$-N concentrations are presented as mean values ($n = 3$) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Day</th>
<th>DBL (cm)</th>
<th>$R^2$</th>
<th>Average grab sample ($\mu$g L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ with DBL ($\mu$g L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ without DBL ($\mu$g L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 - 2</td>
<td>0.095 ± 0.006</td>
<td>0.9989</td>
<td>327.6 ± 69.9</td>
<td>337.1 ± 12.2</td>
<td>164.0 ± 5.9</td>
</tr>
<tr>
<td>2 - 4</td>
<td>0.141 ± 0.065</td>
<td>0.9257</td>
<td>223.4 ± 31.0</td>
<td>215.4 ± 5.7</td>
<td>83.9 ± 2.2</td>
</tr>
<tr>
<td>4 - 6</td>
<td>0.038 ± 0.009</td>
<td>0.9950</td>
<td>211.7 ± 85.5</td>
<td>238.7 ± 8.9</td>
<td>167.8 ± 6.3</td>
</tr>
</tbody>
</table>
Appendix 3. Supporting Information for the research article included as Chapter 3

Development and evaluation of a diffusive gradients in a thin film technique for measuring ammonium in freshwaters

Jianyin Huang\textsuperscript{a}, William W. Bennett\textsuperscript{a}, David T. Welsh\textsuperscript{a}\textsuperscript{*}, Tianling Li\textsuperscript{a}, and Peter R. Teasdale\textsuperscript{a}

\textsuperscript{a} Environmental Futures Research Institute, Griffith University, Gold Coast campus, QLD 4215, Australia

\textsuperscript{*} Corresponding Author: d.welsh@griffith.edu.au. Tel: +61 (07) 555 29186. Fax: +61 (07) 555 28067
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Fig S1. Mass of NH$_4$-N accumulated by PrCH-DGT for 24 h of $\Delta g = 0.09$ cm.

Fig S2. The speciation of NH$_4^+$ (aq), NH$_3$ (aq) and NH$_3$ (g) at pH 1 - 14.

Fig S3. Changes in NH$_4$-N concentration measured in grab samples and PrCH-DGT values with various diffusive gel thicknesses at Mudgeeraba Creek and Loders Creek.

Fig S4. Plot of reciprocal mass of NH$_4$-N accumulated by PrCH-DGT versus diffusive layer thickness at Mudgeeraba Creek and Loders Creek.

Table S1. Grab sample, and $C_{DGT}$ NH$_4$-N concentrations calculated with different thickness diffusive layers at Mudgeeraba Creek for 2- and 4-day deployment.

Table S2. Grab sample, and $C_{DGT}$ NH$_4$-N concentrations calculated with different thickness diffusive layers at Loders Creek for 24 h and 48 h deployment.

Table S3. DBL thicknesses, mean grab sample and $C_{DGT}$ NH$_4$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Mudgeeraba Creek.

Table S4. DBL thicknesses, mean grab sample and $C_{DGT}$ NH$_4$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Loders Creek.

Table S5. DBL calculation at Mudgeeraba Creek.

Table S6. DBL calculation at Loders Creek.
The high $R^2$ value (≥ 0.9960) of NH$_4$-N accumulation confirmed that the new DGT method met the assumption of the DGT equation of linear mass accumulation over 24 h.

**Fig S1.** Mass of NH$_4$-N accumulated by PrCH-DGT for 24 h of $\Delta g = 0.09$ cm. The solid line was the regression line calculated based on the mass of NH$_4$-N accumulated by PrCH-DGT at 4, 8, 12, 16 and 24 h with $R^2 = 0.9960$ and $y = 6.45555x - 6.1927$. Experimental conditions: initial NH$_4$-N concentration = 1.71 mg L$^{-1}$, final NH$_4$-N concentration = 1.54 mg L$^{-1}$; conductivity = 0.354 ± 0.031 mS cm$^{-1}$; temperature = 25.2 ± 0.19 °C.
Approximately 15% of NH₄-N would be present as ammonia at pH 8.53 (Fig S2) – there was no evidence for a low ratio at this pH (Table 1) which suggests that NH₃ is measured by PrCH-DGT. Data in Fig. S2 was obtained using Visual MINTEQ 3.1.

**Fig S2.** The speciation of NH₄⁺(aq) (black), NH₃(aq) (light grey) and NH₃ (g) (long dash) at pH 1-14. The dashed line represents the pH at 8.5.

Fig S3 shows the C_{DGT} values measured with PrCH-DGTs with different diffusive layer thicknesses at Mudgeeraba Creek over 2- and 4- day deployments, and Loders Creek for 24 h and 48 h deployments.
Fig S3. Changes in NH₄-N concentration measured in grab samples (○) and PrCH-DGT values with various diffusive gel thicknesses at Mudgeeraba Creek (A) and Loders Creek (B): 0.05 cm (■), 0.09 cm (◆), 0.13 cm (▲). Data are mean values (n = 3) ± 1 standard deviation.
Table S1. Mean grab sample values, and $C_{\text{DGT}}$ NH$_4$-N concentrations calculated with different thickness diffusive layers at Mudgeeraba Creek for 2- and 4-day deployment. Data for NH$_4$-N concentrations are presented as mean values ($n = 3$) ± 1 standard deviation. These data are corrected for DBL using the average DBL from Tables S3 and S4.

<table>
<thead>
<tr>
<th>Time (d)</th>
<th>Average grab sample (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ (0.05 cm) (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ (0.09 cm) (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ (0.13 cm) (μg L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>43.90 ± 21.08</td>
<td>50.61 ± 2.26</td>
<td>46.66 ± 3.01</td>
<td>47.95 ± 0.76</td>
</tr>
<tr>
<td>4</td>
<td>33.55 ± 21.01</td>
<td>15.11 ± 4.03</td>
<td>25.17 ± 1.32</td>
<td>28.53 ± 2.31</td>
</tr>
</tbody>
</table>

Table S2. Mean grab sample values, and $C_{\text{DGT}}$ NH$_4$-N concentrations calculated with different thickness diffusive layers at Loders Creek 24 h and 48 h deployment. Data for NH$_4$-N concentrations are presented as mean values ($n = 3$) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>Average grab sample (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ (0.05 cm) (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ (0.09 cm) (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ (0.13 cm) (μg L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>24</td>
<td>104.9 ± 78.4</td>
<td>89.83 ± 9.57</td>
<td>119.6 ± 19.4</td>
<td>123.9 ± 4.23</td>
</tr>
<tr>
<td>48</td>
<td>139.9 ± 116.3</td>
<td>109.7 ± 2.81</td>
<td>193.2 ± 10.9</td>
<td>212.4 ± 1.57</td>
</tr>
</tbody>
</table>

Fig S4 showed example plots of 1/mass (ng$^{-1}$) versus Δg (cm) for PrCH-DGT probes used to determine the DBL thickness at each site. The DBL ranged from 0.096 to 0.203 cm at Mudgeeraba Creek and 0.030 to 0.041 cm at Loders Creek, which are approximately 36 - 230% of the standard DGT diffusive layer thickness (Δg = 0.09 cm). Most $C_{\text{DGT}}$ values calculated taking the DBL thickness into consideration provided good matches for the
average NH₄-N concentrations measured in grab water samples collected over the same deployment period (Table S3 and S4).

**Fig S4.** Plot of reciprocal mass of NH₄-N accumulated by PrCH-DGT versus diffusive layer thickness (Δg; cm) for Mudgeeraba Creek on day 7th - 8th Oct and Loders Creek on day 7th Nov 10 am -18pm. Data points are mean values (n = 3) ± 1 standard deviation. Mudgeeraba Creek (●): R² = 0.9998; DBL = 0.203 ± 0.004 cm; pH = 6.87 ± 0.03; conductivity 0.373 ± 0.0045 mS cm⁻¹; temperature = 20.4 ± 0.5 °C. Loders Creek (■): R² = 0.9997; DBL = 0.030 ± 0.002 cm; pH = 6.73 ± 0.04; conductivity = 0.896 ± 0.055 mS cm⁻¹; temperature = 23.0 ± 0.7 °C.
Table S3. DBL thicknesses, and grab sample, and $C_{\text{DGT}}$ NH$_4$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Mudgeeraba Creek. Data for NH$_4$-N concentrations are presented as mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time (d)</th>
<th>DBL (cm)</th>
<th>R$^2$</th>
<th>Average grab sample (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ with DBL (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ without DBL (μg L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0.203 ± 0.004</td>
<td>0.9998</td>
<td>53.5 ± 18.3</td>
<td>60.9 ± 1.4</td>
<td>18.7 ± 0.4</td>
</tr>
<tr>
<td>2</td>
<td>0.118 ± 0.031</td>
<td>0.9826</td>
<td>32.6 ± 11.2</td>
<td>48.9 ± 5.9</td>
<td>21.2 ± 2.6</td>
</tr>
<tr>
<td>3</td>
<td>0.096 ± 0.021</td>
<td>0.9906</td>
<td>18.3 ± 9.1</td>
<td>30.8 ± 2.4</td>
<td>14.9 ± 1.2</td>
</tr>
<tr>
<td>4</td>
<td>0.109 ± 0.041</td>
<td>0.9706</td>
<td>18.0 ± 8.6</td>
<td>24.9 ± 1.4</td>
<td>11.3 ± 0.6</td>
</tr>
</tbody>
</table>

Table S4. DBL thicknesses, and grab sample, and $C_{\text{DGT}}$ NH$_4$-N concentrations calculated with and without inclusion of the DBL for each DGT deployment at Loders Creek. Data for NH$_4$-N concentrations are presented as mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time (h)</th>
<th>DBL (cm)</th>
<th>R$^2$</th>
<th>Average grab sample (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ with DBL (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ without DBL (μg L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>8</td>
<td>0.030 ± 0.002</td>
<td>0.9997</td>
<td>69.0 ± 39.8</td>
<td>68.2 ± 4.0</td>
<td>51.2 ± 3.0</td>
</tr>
<tr>
<td>16</td>
<td>0.041 ± 0.008</td>
<td>0.9991</td>
<td>145.9 ± 37.8</td>
<td>154.8 ± 20.6</td>
<td>106.4 ± 14.2</td>
</tr>
<tr>
<td>8</td>
<td>0.039 ± 0.022</td>
<td>0.9806</td>
<td>184.8 ± 8.2</td>
<td>175.4 ± 11.6</td>
<td>122.4 ± 8.1</td>
</tr>
<tr>
<td>16</td>
<td>0.034 ± 0.003</td>
<td>0.9995</td>
<td>264.9 ± 87.3</td>
<td>329.4 ± 15.3</td>
<td>239.1 ± 11.1</td>
</tr>
</tbody>
</table>

Tables S5 and S6 show the use of R$^2$ and the equation from DBL plots (as shown in Fig S4) to validate field DGT deployments. This is appropriate as the use of different diffusive layer thickness over the same time is equivalent to the mass vs. time plot (Fig S1) in determining
whether the data is consistent with the DGT equation. The data in italics uses the data from Fig S3. The rest of the data is from either Table S3 or S4. The first line in each table is from the data in Fig S4.

The results in bold can be considered to have failed the field validation. The 4-day deployment at Mudgeeraba creek fails because of the negative slope in the equation. The fact that a high $R^2$ value was obtained is curious and may be because the measurements at the different diffusive layer thickness were significantly different. This clearly indicates that the DGTs with thinner diffusive layers had exceeded the limit of linear accumulation for the deployment conditions. The 48 h data at Loders creek fails because the $R^2$ value is very low and the slope indicates the curve is very flat. This also suggests that the mass accumulated in the thinner diffusive gels had exceeded the limit of linear accumulation for the deployment conditions.

The other results in italics have lower $R^2$ values and for the 24 hour data at Loders Creek the 0.05 cm diffusive layer was significantly different. This is consistent with these deployment conditions being selected to test the boundaries of quantitative DGT measurements, whereas the non-italic deployment conditions were selected to ensure quantitative results. As the results in Fig 5 suggest, there is no way to predict the limits of linear accumulation for any given deployment conditions so the use of DBL measurement data in this way is suggested as a useful field validation. On the basis of these results we recommend that an $R^2$ value of 0.95 be considered an acceptable result for a field validation of the PrCH-DGT for the measurement of NH$_4$-N concentrations, as long as the slope of the equation is positive.
Table S5. DBL calculation at Mudgeeraba Creek.

<table>
<thead>
<tr>
<th>Deployment date</th>
<th>Deployment time (d)</th>
<th>$R^2$</th>
<th>Equation</th>
</tr>
</thead>
<tbody>
<tr>
<td>7th - 8th Oct</td>
<td>1</td>
<td>0.9998</td>
<td>0.000873x + 0.000201</td>
</tr>
<tr>
<td>8th - 9th Oct</td>
<td>1</td>
<td>0.9826</td>
<td>0.002894x + 0.000137</td>
</tr>
<tr>
<td>9th - 10th Oct</td>
<td>1</td>
<td>0.9906</td>
<td>0.002397x + 0.000189</td>
</tr>
<tr>
<td>10th - 11th Oct</td>
<td>1</td>
<td>0.9706</td>
<td>0.002294x + 0.000201</td>
</tr>
<tr>
<td>7th - 9th Oct</td>
<td>2</td>
<td>0.9375</td>
<td>0.000818x + 0.000105</td>
</tr>
<tr>
<td>7th - 11th Oct</td>
<td>4</td>
<td>0.9369</td>
<td>-0.004104x + 0.000858</td>
</tr>
</tbody>
</table>

Table S6. DBL calculation at Loders Creek.

<table>
<thead>
<tr>
<th>Deployment date</th>
<th>Deployment time (h)</th>
<th>$R^2$</th>
<th>Equation</th>
</tr>
</thead>
<tbody>
<tr>
<td>7th Nov 10 am - 18 pm</td>
<td>8</td>
<td>0.9997</td>
<td>0.003052x + 0.000144</td>
</tr>
<tr>
<td>7th 18 pm - 8th Nov 10 am</td>
<td>16</td>
<td>0.9991</td>
<td>0.000696x + 0.000026</td>
</tr>
<tr>
<td>8th Nov 10 am - 18 pm</td>
<td>8</td>
<td>0.9806</td>
<td>0.001039x + 0.000034</td>
</tr>
<tr>
<td>8th 18 pm - 9th Nov 10 am</td>
<td>16</td>
<td>0.9995</td>
<td>0.001079x + 0.000028</td>
</tr>
<tr>
<td>7th - 8th Nov</td>
<td>24</td>
<td>0.9337</td>
<td>0.000508x + 0.000050</td>
</tr>
<tr>
<td>7th - 9th Nov</td>
<td>48</td>
<td>0.075</td>
<td>0.000006x + 0.000036</td>
</tr>
</tbody>
</table>
Appendix 4. Supporting Information for the research article included as Chapter 4

Determining time-weighted average concentrations of nitrate and ammonium in freshwaters using DGT with ion exchange membrane-based binding layers

Jianyin Huang\textsuperscript{a}, William W. Bennett\textsuperscript{a}, David T. Welsh\textsuperscript{a*}, and Peter R. Teasdale\textsuperscript{a}

\textsuperscript{a} Environmental Futures Research Institute, Griffith University, Gold Coast campus, QLD 4215, Australia

* Corresponding Author: d.welsh@griffith.edu.au. Tel: +61 (07) 555 29186. Fax: +61 (07) 555 28067
Contents

**Figure S1.** Mass of NH$_4$-N accumulated by CMI-DGT over 24 h at various ionic strength solutions.

**Figure S2.** Relationship between the diffusion coefficient of NH$_4$-N and conductivity for CMI-DGT.

**Figure S3.** Capacity of AMI-DGT for NO$_3$-N and CMI-DGT for NH$_4$-N.

**Table S1.** Comparison of the uptake and elution efficiencies for binding membranes and hydrogels.

**Table S2.** Comparison of uptake of binding membranes and hydrogels for different pH.

**Table S3.** NO$_3$-N concentrations at Loders Creek: $C_{\text{SOLN}}$, $C_{\text{A520E-DGT}}$ and $C_{\text{AMI-DGT}}$.

**Table S4.** NH$_4$-N concentrations at Loders Creek: $C_{\text{SOLN}}$, $C_{\text{PrCH-DGT}}$ and $C_{\text{CMI-DGT}}$.

**Table S5.** NO$_3$-N concentrations at Saltwater Creek: $C_{\text{SOLN}}$, $C_{\text{A520E-DGT}}$ and $C_{\text{AMI-DGT}}$.

**Table S6.** NH$_4$-N concentrations at Saltwater Creek: $C_{\text{SOLN}}$, $C_{\text{PrCH-DGT}}$ and $C_{\text{CMI-DGT}}$. 
AMI and CMI membranes had high uptake and elution efficiencies for NO₃⁻-N and NH₄⁺-N, respectively.

**Table S1.** Comparison of the uptake and elution efficiencies for binding membranes and hydrogels.

<table>
<thead>
<tr>
<th>Binding agents</th>
<th>AMI</th>
<th>A520E¹</th>
<th>CMI</th>
<th>PrCH²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uptake efficiency (%)</td>
<td>96.7 ± 3.3</td>
<td>98.7 ± 1.4</td>
<td>94.8 ± 2.3</td>
<td>92.5 ± 5.1</td>
</tr>
<tr>
<td>Elution efficiency (%)</td>
<td>77.6 ± 6.1</td>
<td>82.7 ± 4.2</td>
<td>89.9 ± 4.6</td>
<td>87.2 ± 5.3</td>
</tr>
</tbody>
</table>

The ratios of $C_{\text{AMI-DGT}}:C_{\text{SOLN}}$ and $C_{\text{CMI-DGT}}:C_{\text{SOLN}}$ were between 0.87 and 1.06, suggesting that the binding layer membranes produced similar results to the resin binding gels.

**Table S2.** Comparison of the uptake of binding membranes and hydrogels for different pH.

<table>
<thead>
<tr>
<th>pH</th>
<th>$C_{\text{AMI-DGT}}/C_{\text{SOLN}}$</th>
<th>$C_{\text{A520E-DGT}}/C_{\text{SOLN}}$</th>
<th>$C_{\text{CMI-DGT}}/C_{\text{SOLN}}$</th>
<th>$C_{\text{PrCH-DGT}}/C_{\text{SOLN}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>3.5</td>
<td>0.87 ± 0.02</td>
<td>0.94 ± 0.04</td>
<td>0.89 ± 0.07</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>0.91 ± 0.06</td>
<td>0.92 ± 0.11</td>
<td>0.95 ± 0.07</td>
</tr>
<tr>
<td></td>
<td>7</td>
<td>1.01 ± 0.03</td>
<td>1.00 ± 0.02</td>
<td>0.91 ± 0.03</td>
</tr>
<tr>
<td></td>
<td>8.5</td>
<td>0.93 ± 0.06</td>
<td>0.89 ± 0.03</td>
<td>0.90 ± 0.03</td>
</tr>
</tbody>
</table>

Similar NH₄⁺ diffusion coefficients were found in CMI-DGT and PrCH-DGT.² Mass of accumulated NH₄⁻-N on CMI-DGT decreased with increasing conductivity (Figure S3). High diffusion coefficient of NH₄⁺ was found at low ionic strength (conductivity) and decreased with increasing conductivity (Figure S2).
Figure S1. Mass of NH$_4$-N accumulated by CMI-DGT over 24 h at various ionic strength solutions. The solid line is a regression line. Experimental conditions: NH$_4$-N = 1.5 ± 0.21 mg L$^{-1}$; conductivity = 0.0855 ± 0.001 mS cm$^{-1}$ (■), 0.203 ± 0.029 mS cm$^{-1}$ (○); 0.469 ± 0.019 mS cm$^{-1}$ (▲); 0.683 ± 0.016 mS cm$^{-1}$ (▽); 1.0 ± 0.023 mS cm$^{-1}$ (★).
**Figure S2.** Relationship between the diffusion coefficient of NH$_4$-N and conductivity for CMI-DGT (△) and PrCH-DGT (▼) using an agarose diffusive layer. Data were modified according to the Stokes-Einstein equation to 25 °C. pH = 7.07 ± 0.44. The trend line for the exponential relationship between diffusion coefficient ($D$) and conductivity for CMI-DGT was $R^2 = 0.9979$, $D = 0.000115$ Conductivity$^{-0.346833}$ and PrCH-DGT was $R^2 = 0.9918$, $D = 0.000227$ Conductivity$^{-0.432705}$. 
Membrane DGTs have high binding capacities for NO$_3$-N (921 ± 88 μg) and NH$_4$-N (3512 ± 51 μg).

**Figure S3.** (A) Binding capacity of AMI-DGT (■) for NO$_3$-N and (B) CMI-DGT (▲) for NH$_4$-N. Data are mean values (n = 3) ± 1 standard deviation. Experimental conditions: (A) initial NO$_3$-N = 13.6 mg L$^{-1}$, final NO$_3$-N = 12.3 μg L$^{-1}$; temperature = 24.1 ± 0.5 °C; (B) initial NH$_4$-N = 33.1 mg L$^{-1}$, final NH$_4$-N = 30.6 mg L$^{-1}$; temperature = 23.5 ± 0.4 °C; both conductivities are between 0.41 - 0.45 mS cm$^{-1}$. 
### Table S3. NO$_3$-N concentrations (μg L$^{-1}$) at Loders Creek: $C_{\text{SOLN}}$, $C_{\text{A520E-DGT}}$ and $C_{\text{AMI-DGT}}$.  
Concentrations are presented as mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time</th>
<th>$C_{\text{SOLN}}$</th>
<th>$C_{\text{AMI-DGT}}$</th>
<th>$C_{\text{A520E-DGT}}$</th>
<th>$C_{\text{AMI-DGT:CA520E-DGT}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>8 - 11 Dec</td>
<td>320.4 ± 205.0</td>
<td>404.1 ± 17.3</td>
<td>410.3 ± 6.0</td>
<td>0.98 ± 0.04</td>
</tr>
<tr>
<td>11 - 14 Dec</td>
<td>303.4 ± 74.9</td>
<td>268.5 ± 7.4</td>
<td>285.6 ± 1.6</td>
<td>0.94 ± 0.03</td>
</tr>
<tr>
<td>14 - 17 Dec</td>
<td>180.7 ± 43.3</td>
<td>178.5 ± 2.9</td>
<td>184.4 ± 2.1</td>
<td>0.97 ± 0.01</td>
</tr>
</tbody>
</table>

### Table S4. NH$_4$-N concentrations (μg L$^{-1}$) at Loders Creek: $C_{\text{SOLN}}$, $C_{\text{PCH-DGT}}$ and $C_{\text{CMI-DGT}}$.  
Concentrations are presented as mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time</th>
<th>$C_{\text{SOLN}}$</th>
<th>$C_{\text{CMI-DGT}}$</th>
<th>$C_{\text{PCH-DGT}}$</th>
<th>$C_{\text{CMI-DGT:PCH-DGT}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>8 - 11 Dec</td>
<td>167.5 ± 70.6</td>
<td>217.0 ± 12.4</td>
<td>238.0 ± 13.0</td>
<td>0.91 ± 0.08</td>
</tr>
<tr>
<td>11 - 14 Dec</td>
<td>217.2 ± 68.0</td>
<td>192.3 ± 0.7</td>
<td>229.6 ± 25.9</td>
<td>0.85 ± 0.09</td>
</tr>
<tr>
<td>14 - 17 Dec</td>
<td>269.1 ± 119.1</td>
<td>279.8 ± 20.7</td>
<td>309.9 ± 6.9</td>
<td>0.90 ± 0.07</td>
</tr>
</tbody>
</table>

### Table S5. NO$_3$-N concentrations (μg L$^{-1}$) at Saltwater Creek: $C_{\text{SOLN}}$, $C_{\text{A520E-DGT}}$ and $C_{\text{AMI-DGT}}$.  
Concentrations are presented as mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time</th>
<th>$C_{\text{SOLN}}$</th>
<th>$C_{\text{AMI-DGT}}$</th>
<th>$C_{\text{A520E-DGT}}$</th>
<th>$C_{\text{AMI-DGT:CA520E-DGT}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>27 - 30 Jan</td>
<td>209.6 ± 58.5</td>
<td>256.8 ± 5.1</td>
<td>257.4 ± 8.0</td>
<td>1.06 ± 0.04</td>
</tr>
<tr>
<td>31 Jan - 2 Feb</td>
<td>136.2 ± 19.9</td>
<td>159.2 ± 15.7</td>
<td>150.9 ± 9.5</td>
<td>1.06 ± 0.06</td>
</tr>
<tr>
<td>2 - 5 Feb</td>
<td>181.4 ± 32.7</td>
<td>184.3 ± 17.6</td>
<td>176.2 ± 16.2</td>
<td>1.10 ± 0.24</td>
</tr>
</tbody>
</table>
**Table S6.** NH$_4$-N concentrations (μg L$^{-1}$) at Saltwater Creek: $C_{\text{SOLN}}$, $C_{\text{PrCH-DGT}}$ and $C_{\text{CMI-DGT}}$.

Concentrations are presented as mean values ($n = 3$) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time</th>
<th>$C_{\text{SOLN}}$</th>
<th>$C_{\text{CMI-DGT}}$</th>
<th>$C_{\text{PrCH-DGT}}$</th>
<th>$C_{\text{CMI-DGT}, C_{\text{PrCH-DGT}}}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>27 - 30 Jan</td>
<td>22.7 ± 5.4</td>
<td>20.8 ± 3.3</td>
<td>20.2 ± 1.3</td>
<td>1.11 ± 0.15</td>
</tr>
<tr>
<td>31 Jan - 2</td>
<td>17.0 ± 4.7</td>
<td>15.2 ± 0.2</td>
<td>13.7 ± 1.3</td>
<td>1.11 ± 0.09</td>
</tr>
<tr>
<td>Feb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 - 5 Feb</td>
<td>15.2 ± 5.6</td>
<td>16.5 ± 1.9</td>
<td>14.1 ± 1.6</td>
<td>1.21 ± 0.09</td>
</tr>
</tbody>
</table>

**References**


Appendix 5. Supporting Information for the research article included as Chapter 5

Diffusive gradients in thin films techniques provide representative time-weighted average measurements of inorganic nutrients in dynamic freshwater systems

Jianyin Huang\textsuperscript{a}, William W. Bennett\textsuperscript{a}, David T. Welsh\textsuperscript{a}\textsuperscript{*}, Tianling Li\textsuperscript{a} and Peter R. Teasdal\textsuperscript{a}

\textsuperscript{a} Environmental Futures Research Institute, Griffith University, Gold Coast campus, QLD 4215, Australia

* Corresponding Author: d.welsh@griffith.edu.au. Tel: +61 (07) 555 29186. Fax: +61 (07) 555 28067
Contents

Experiments

Preparation of Metsorb binding layers; Variability of physicochemical parameters; Diffusion boundary layer (DBL) calculations and corrections; Grab sample measurements; Time series deployment at Currumbin Creek.

Results

Figure S1. Map for the field sites.

Figure S2. Maps for the field sites in detail.

Figure S3. The deployment apparatus for DGT samplers.

Figure S4. The influence of rainfall on conductivities at Saltwater Creek and Saltwater Creek pond.

Figure S5. Plot of reciprocal mass of PO$_4$-P (ng$^{-1}$) accumulated by Metsorb-DGT or NO$_3$-N (ng$^{-1}$) by A520E-DGT versus diffusive layer thickness (Δg; cm) at different sites.

Figure S6. Grab samples of NH$_4$-N, NO$_3$-N and PO$_4$-P at Saltwater Creek, Botanic Gardens and Currumbin Creek, and the rainfall events.

Table S1. The mean, standard deviation (SD) and relative standard deviation (RSD) of pH, temperature, conductivity and dissolved oxygen at each field site.

Table S2. DO (%) in the early morning and afternoon at each site.

Table S3. Measured DBL thickness at each site for 24 h and 72 h deployments.

Table S4. NH$_4$-N concentrations (μg L$^{-1}$) in each site: $C_{\text{SOLN}}$ and $C_{\text{PCH-DGT}}$ with and without DBL.

Table S5. NO$_3$-N concentrations (μg L$^{-1}$) in each site: $C_{\text{SOLN}}$ and $C_{\text{A520E-DGT}}$ with and without DBL.
Table S6. PO₄-P concentrations (μg L⁻¹) in each site: C_SOLN and C_Metsorb-DGT with and without DBL.

Table S7. DIN and PO₄-P concentrations (μg L⁻¹) at Currumbin Creek.

**Preparation of Metsorb binding layers.** Metsorb binding layers were prepared in a fume hood. 1 g of TiO₂ powder was added to 10 mL of gel solution (18.75 mL of 15% acrylamide, 7.5 g of 0.3% cross-linker and 23.75 mL of deionised water). The solution was sonicated for approximately 5 minutes to ensure the TiO₂ powder was suspended in the solution. 60 μL of freshly prepared 10% ammonium persulfate solution and 20 μL of N,N,N’,N’-tetramethyl ethylenediamine (TEMED) were added while stirring. The solution was cast immediately between glass plated separated by a 0.25 mm thick plastic spacer and set at 45 °C for 1 h. After that, the binding layers were washed 2 - 3 times before being stored in deionised water at 4 °C.

**Variability of physicochemical parameters.** Our results show that pH, conductivity, temperature and dissolved oxygen (DO) concentrations (% saturation) often differ among the seven sites and some are highly variable over time (Table S1). Conductivities at Saltwater Creek and Saltwater Creek pond were high when there was no rain and declined during rainfall. After the rain stopped, conductivities began to increase (Figure S4). These results suggest that groundwater in the Saltwater Creek catchment may have a slight salinity.
Figure S1. Map for the field sites
Figure S2. Maps for the field sites in detail
Figure S3. The deployment apparatus for DGT samplers.

Table S1. The mean, standard deviation (SD) and relative standard deviation (RSD) of pH, conductivity, temperature and dissolved saturation oxygen at each field site.

<table>
<thead>
<tr>
<th>Field site</th>
<th>pH</th>
<th>Temperature (°C)</th>
<th>Conductivity (mS cm⁻¹)</th>
<th>Dissolved saturation oxygen (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SD</td>
<td>RSD</td>
<td>Mean</td>
</tr>
<tr>
<td>Saltwater Creek</td>
<td>7.47</td>
<td>0.07</td>
<td>0.9</td>
<td>17.4</td>
</tr>
<tr>
<td>Saltwater Creek pond</td>
<td>7.24</td>
<td>0.25</td>
<td>3.5</td>
<td>19.3</td>
</tr>
<tr>
<td>Loders Creek</td>
<td>6.91</td>
<td>0.13</td>
<td>1.9</td>
<td>16.3</td>
</tr>
<tr>
<td>Botanic Gardens</td>
<td>7.23</td>
<td>0.09</td>
<td>1.2</td>
<td>17.5</td>
</tr>
<tr>
<td>Worongary Creek</td>
<td>6.61</td>
<td>0.05</td>
<td>0.8</td>
<td>17.4</td>
</tr>
<tr>
<td>Currumbin Creek</td>
<td>7.29</td>
<td>0.12</td>
<td>1.6</td>
<td>14.4</td>
</tr>
<tr>
<td>Currumbin Creek wetland</td>
<td>6.90</td>
<td>0.07</td>
<td>1.0</td>
<td>15.8</td>
</tr>
</tbody>
</table>
Figure S4. The influence of rainfall on conductivities at Saltwater Creek and Saltwater Creek pond.

Changes in dissolved oxygen (DO) concentrations (% saturation) were investigated with respect to time of analysis. The results showed that the DO in the afternoon was generally higher than that in the early morning. The results showed that the DO in the afternoon was generally higher than that in the early morning. This is consistent with the typical pattern of biological productivity in freshwater streams, with respiration dominating at night and photosynthesis during the day. Many of the freshwater systems are under-saturated with respect to DO, except for Currumbin Creek stream.
Table S2. DO (%) in the early morning and afternoon at each site. M = morning and A = afternoon.

<table>
<thead>
<tr>
<th>Field site</th>
<th>Day 1 M</th>
<th>Day 1 A</th>
<th>Day 2 M</th>
<th>Day 2 A</th>
<th>Day 3 M</th>
<th>Day 3 A</th>
<th>Day 4 M</th>
<th>Day 4 A</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saltwater Creek</td>
<td>41</td>
<td>58</td>
<td>44</td>
<td>54.3</td>
<td>48</td>
<td>55</td>
<td>43</td>
<td></td>
</tr>
<tr>
<td>Saltwater Creek pond</td>
<td>54</td>
<td>60</td>
<td>55</td>
<td>63</td>
<td>51</td>
<td>70</td>
<td>48</td>
<td></td>
</tr>
<tr>
<td>Loders Creek</td>
<td>-</td>
<td>43.5</td>
<td>39</td>
<td>43</td>
<td>36.8</td>
<td>67</td>
<td>62.7</td>
<td>54.8</td>
</tr>
<tr>
<td>Botanic Gardens</td>
<td>-</td>
<td>81</td>
<td>56.9</td>
<td>75.9</td>
<td>63.3</td>
<td>72.9</td>
<td>75.5</td>
<td>82.1</td>
</tr>
<tr>
<td>Worongary Creek</td>
<td>-</td>
<td>83.1</td>
<td>72</td>
<td>79.3</td>
<td>71.8</td>
<td>73.1</td>
<td>70.4</td>
<td>76.8</td>
</tr>
<tr>
<td>Currumbin Creek</td>
<td>-</td>
<td>101.6</td>
<td>103</td>
<td>97</td>
<td>95.4</td>
<td>106.3</td>
<td>110</td>
<td>104</td>
</tr>
<tr>
<td>Currumbin Creek wetland</td>
<td>-</td>
<td>52.2</td>
<td>71.7</td>
<td>48.3</td>
<td>76.1</td>
<td>80.3</td>
<td>69.2</td>
<td>76</td>
</tr>
</tbody>
</table>

**Diffusive boundary layer (DBL) calculations and corrections.** Previous field deployments in this study\(^1\), \(^2\) have demonstrated the importance of measuring the DBL thickness and correcting the DGT calculation. The thicknesses of the DBL were calculated from the slope and y-intercept of the plots according to previous studies\(^3\), \(^4\). Figure S5 shows DBL plots have good linearity (R\(^2\) > 0.94) with relative standard deviation less than 20%, indicating that the technique is reproducible.
Figure S5. Plot of reciprocal mass of \( \text{PO}_4 \)-P accumulated (ng\(^{-1}\)) by Metsorb-DGT or NO\(_3\)-N (ng\(^{-1}\)) by A520E-DGT versus diffusive layer thickness (\( \Delta g \); cm) at different field sites. Saltwater Creek (A): R\(^2\) = 0.9991, DBL = 0.045 ± 0.005 cm; Saltwater Creek Pond (B): R\(^2\) = 0.9571, DBL = 0.088 ± 0.0045 cm; Loders Creek (C): R\(^2\) = 0.9912, DBL = 0.027 ± 0.013 cm; Botanic Gardens (D): R\(^2\) = 0.9455, DBL = 0.101 ± 0.054 cm; Currumbin Creek (E): R\(^2\) = 0.9996, DBL = 0.036 ± 0.003 cm.
Determination of the DBL thickness was done using PO₄-P and NO₃-N measurements, however, DBLs were not obtained for all sites. Although the results showed that DBL thicknesses varied among field sites, some trends were apparent (Table S3). The wetland sites, such as at Saltwater Creek pond and the Botanic Gardens, tended to have similar high DBL thicknesses (0.085 - 0.101 cm), whereas the streams (Saltwater Creek, Loders Creek and Currumbin Creek) had similar and lower DBL thicknesses (0.021 - 0.059 cm). This is consistent with previous observations.¹ ² Importantly, our data indicates that the DBL thicknesses from field sites with similar hydrodynamic conditions are quite similar. Therefore, the average DBL from Saltwater Creek and the Botanic Gardens wetlands (0.0925 cm) was used at Currumbin Creek wetlands site and the average DBL from Saltwater Creek, Loders Creek and Currumbin Creek streams (0.0366 cm) was applied at Worongary Creek.

**Table S3.** Measured DBL thicknesses at each field site for 24 h and 72 h deployments.

<table>
<thead>
<tr>
<th>Field site</th>
<th>R²</th>
<th>DBL (cm) at 24 h</th>
<th>R²</th>
<th>DBL (cm) at 72 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saltwater Creek</td>
<td>-</td>
<td>-</td>
<td>0.9991</td>
<td>0.045 ± 0.005</td>
</tr>
<tr>
<td>Saltwater Creek pond</td>
<td>0.9691</td>
<td>0.085 ± 0.035</td>
<td>0.9571</td>
<td>0.088 ± 0.045</td>
</tr>
<tr>
<td>Loders Creek</td>
<td>0.9771</td>
<td>0.021 ± 0.019</td>
<td>0.9912</td>
<td>0.027 ± 0.013</td>
</tr>
<tr>
<td>Botanic Gardens</td>
<td>0.9881</td>
<td>0.096 ± 0.023</td>
<td>0.9455</td>
<td>0.101 ± 0.054</td>
</tr>
<tr>
<td>Currumbin Creek</td>
<td>0.9996</td>
<td>0.036 ± 0.003</td>
<td>0.9608</td>
<td>0.054 ± 0.033</td>
</tr>
</tbody>
</table>

The results demonstrated that the DBL can have a significant impact on DGT measurements (Tables S4 - S6). The DBL thicknesses observed at the wetland sites are approximately 100% of the standard DGT diffusive layer thickness (Δg = 0.09 cm) and therefore introduce a substantial error into the final concentration determined. Even the DBL present in flowing streams can lead to underestimation of the concentrations determined. Cᵢ values calculated
with the DBL included agree better with the average values of grab samples collected over the same deployment time in many instances. For example, at the Saltwater Creek Pond, DGT-measured concentrations of NH$_4$-N, NO$_3$-N and PO$_4$-P with the DBL thickness accounted for are 107%, 97.2% and 114.9% of the average concentrations measured in grab samples, respectively, while DGT-measured concentrations assuming no DBL are only 55%, 48.6% and 58.1% of the average values of grab samples, respectively. As a result, it is recommended that DBL thicknesses are routinely estimated for field measurements of nutrients using the DGT techniques.

**Figure S6.** Mean values of NO$_3$-N (●), NH$_4$-N (■) and PO$_4$-P (▼) in grab samples collected at Saltwater Creek (A), Botanic Gardens (B) and Currumbin Creek (C).
**Table S4.** Average grab sample concentrations of NH$_4$-N, and DGT-measured concentrations of NH$_4$-N ($C_{\text{DGT}}$) with and without the DBL-thickness correction. Data are mean values ($n = 3$) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Field site</th>
<th>Time (h)</th>
<th>Average grab sample (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ without DBL (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ with DBL (μg L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saltwater Creek</td>
<td>72</td>
<td>7.01 ± 2.71</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Saltwater Creek pond</td>
<td>24</td>
<td>51.9 ± 8.34</td>
<td>28.6 ± 3.18</td>
<td><strong>55.3 ± 6.16</strong></td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>67.4 ± 20.3</td>
<td>29.7 ± 4.29</td>
<td><strong>58.1 ± 8.39</strong></td>
</tr>
<tr>
<td>Loders Creek</td>
<td>24</td>
<td>206.7 ± 68.8</td>
<td>200.0 ± 23.7</td>
<td><strong>246.8 ± 29.3</strong></td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>516.1 ± 460.6</td>
<td>437.2 ± 16.7</td>
<td><strong>537.3 ± 20.9</strong></td>
</tr>
<tr>
<td>Botanic Garden</td>
<td>24</td>
<td>24.4 ± 11.4</td>
<td>19.3 ± 3.84</td>
<td><strong>39.8 ± 7.91</strong></td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>25.3 ± 11.3</td>
<td>35.6 ± 4.16</td>
<td><strong>55.1 ± 6.49</strong></td>
</tr>
<tr>
<td>Worongary Creek</td>
<td>72</td>
<td>13.3 ± 2.19</td>
<td>11.2 ± 0.25</td>
<td><strong>14.1 ± 0.38</strong></td>
</tr>
<tr>
<td>Currumbin Creek</td>
<td>24</td>
<td>11.4 ± 3.47</td>
<td>11.6 ± 2.43</td>
<td><strong>16.4 ± 3.56</strong></td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>10.3 ± 2.85</td>
<td>10.4 ± 2.61</td>
<td><strong>14.5 ± 4.65</strong></td>
</tr>
<tr>
<td>Currumbin Creek wetland</td>
<td>72</td>
<td>48.9 ± 32.3</td>
<td>72.0 ± 4.92</td>
<td><strong>104.3 ± 2.13</strong></td>
</tr>
</tbody>
</table>

Queensland Water Guideline for NH$_4$-N is 20 μg L$^{-1}$ in lowland streams and 10 μg L$^{-1}$ in freshwater lakes. The data of $C_{\text{DGT}}$ with DBL in bold is higher than the guideline.
Table S5. Average grab sample concentrations of NO$_3$-N, and DGT-measured concentrations of NO$_3$-N ($C_{\text{DGT}}$) with and without the DBL-thickness correction. Data are mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Field site</th>
<th>Time (h)</th>
<th>Average grab sample (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ without DBL (μg L$^{-1}$)</th>
<th>$C_{\text{DGT}}$ with DBL (μg L$^{-1}$)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saltwater Creek</td>
<td>72</td>
<td>106.3 ± 46.9</td>
<td>113.6 ± 6.31</td>
<td>153.0 ± 8.50</td>
</tr>
<tr>
<td>Saltwater Creek pond</td>
<td>24</td>
<td>83.0 ± 10.1</td>
<td>40.3 ± 3.69</td>
<td>80.7 ± 7.14</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>98.1 ± 18.5</td>
<td>56.0 ± 3.81</td>
<td>109.6 ± 7.46</td>
</tr>
<tr>
<td>Loders Creek</td>
<td>24</td>
<td>183.8 ± 61.5</td>
<td>178.4 ± 4.25</td>
<td>220.1 ± 5.25</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>318.5 ± 131.2</td>
<td>309.4 ± 19.1</td>
<td>373.6 ± 23.0</td>
</tr>
<tr>
<td>Botanic Garden</td>
<td>24</td>
<td>38.7 ± 7.19</td>
<td>24.7 ± 0.47</td>
<td>51.0 ± 0.98</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>37.0 ± 11.9</td>
<td>32.2 ± 0.87</td>
<td>54.0 ± 1.18</td>
</tr>
<tr>
<td>Worongary Creek</td>
<td>72</td>
<td>165.9 ± 13.2</td>
<td>112.8 ± 4.75</td>
<td>144.5 ± 6.09</td>
</tr>
<tr>
<td>Currumbin Creek</td>
<td>24</td>
<td>78.3 ± 2.91</td>
<td>53.4 ± 8.79</td>
<td>75.4 ± 12.4</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>73.7 ± 13.4</td>
<td>54.0 ± 0.26</td>
<td>86.4 ± 0.42</td>
</tr>
<tr>
<td>Currumbin Creek wetland</td>
<td>72</td>
<td>34.6 ± 10.1</td>
<td>27.0 ± 2.47</td>
<td>46.2 ± 4.22</td>
</tr>
</tbody>
</table>

Queensland Water Guideline for NO$_3$-N is 60 μg L$^{-1}$ in lowland streams and 10 μg L$^{-1}$ in freshwater lakes. The data of $C_{\text{DGT}}$ with DBL in bold is higher than the guideline.
Table S6. Average grab sample concentrations of PO₄-P, and DGT-measured concentrations of PO₄-P ($C_{\text{DGT}}$) with and without the DBL-thickness correction. Data are mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Field site</th>
<th>Time(h)</th>
<th>Average grab sample (μg L⁻¹)</th>
<th>$C_{\text{DGT}}$ with DBL (μg L⁻¹)</th>
<th>$C_{\text{DGT}}$ without DBL (μg L⁻¹)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Saltwater Creek</td>
<td>72</td>
<td>9.47 ± 0.66</td>
<td>6.94 ± 0.74</td>
<td>10.4 ± 1.10</td>
</tr>
<tr>
<td>Saltwater Creek pond</td>
<td>24</td>
<td>7.47 ± 0.87</td>
<td>4.34 ± 0.43</td>
<td>8.58 ± 0.86</td>
</tr>
<tr>
<td>Loders Creek</td>
<td>24</td>
<td>10.0 ± 0.97</td>
<td>6.94 ± 0.69</td>
<td>8.56 ± 0.95</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>7.75 ± 2.29</td>
<td>4.57 ± 0.85</td>
<td>5.94 ± 1.10</td>
</tr>
<tr>
<td>Botanic Garden</td>
<td>24</td>
<td>25.3 ± 1.41</td>
<td>12.4 ± 1.34</td>
<td>24.7 ± 2.66</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>26.1 ± 2.17</td>
<td>13.5 ± 1.73</td>
<td>30.6 ± 3.68</td>
</tr>
<tr>
<td>Worongary Creek</td>
<td>72</td>
<td>3.37 ± 0.54</td>
<td>2.00 ± 0.61</td>
<td>2.76 ± 0.87</td>
</tr>
<tr>
<td>Currumbin Creek</td>
<td>24</td>
<td>11.8 ± 0.78</td>
<td>8.84 ± 0.60</td>
<td>12.5 ± 0.84</td>
</tr>
<tr>
<td></td>
<td>72</td>
<td>11.9 ± 1.62</td>
<td>4.81 ± 0.63</td>
<td>7.70 ± 1.00</td>
</tr>
<tr>
<td>Currumbin Creek wetland</td>
<td>72</td>
<td>10.7 ± 1.91</td>
<td>3.16 ± 0.90</td>
<td>11.1 ± 1.96</td>
</tr>
</tbody>
</table>

Queensland Water Guidelines for PO₄-P is 20 µg L⁻¹ in lowland streams and 5 µg L⁻¹ in freshwater lakes. The data of $C_{\text{DGT}}$ with DBL in bold is higher than the guideline.

**Grab sample measurements.** The dissolved inorganic nutrient concentrations at the seven sites are shown in Figures 2 and S6. At Saltwater Creek, NO₃-N concentrations increased from 60 µg L⁻¹ to 145 µg L⁻¹ during the days with rainfall while NH₄-N and PO₄-P concentrations were consistent, indicating NO₃-N concentrations were influenced by run-off. For the Botanic Garden site, the NH₄-N concentrations were higher in the morning (about 37 µg L⁻¹) and lower in the afternoon (15 µg L⁻¹) during the entire deployment time. NO₃-N had
the same pattern, 37 - 50 µg L\(^{-1}\) in the morning and 17 - 39 µg L\(^{-1}\) in the afternoon. Compared with NH\(_4\)-N and NO\(_3\)-N, the fluctuation of PO\(_4\)-P was much smaller, which was between 24 µg L\(^{-1}\) and 27 µg L\(^{-1}\). This is due to the influence of diurnal shifts of biological productivity. At Currumbin Creek, relatively high NO\(_3\)-N concentrations were observed, and NH\(_4\)-N and PO\(_4\)-P concentrations changed slightly from 8 µg L\(^{-1}\) to 13 µg L\(^{-1}\). The high NO\(_3\)-N concentrations could be related to the usage of fertiliser from agricultural lands near the sampling site.

**Time series deployment at Currumbin Creek.** The raw data are shown in Table S7.

**Table S7.** DIN and PO\(_4\)-P concentrations (µg L\(^{-1}\)) at Currumbin Creek. Data are mean values (n = 3) ± 1 standard deviation.

<table>
<thead>
<tr>
<th>Time</th>
<th>NO(_3)-N (µg L(^{-1}))</th>
<th>NH(_4)-N (µg L(^{-1}))</th>
<th>PO(_4)-P (µg L(^{-1}))</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(C_{\text{SOLN}})</td>
<td>(C_{\text{DGT}})</td>
<td>(C_{\text{SOLN}})</td>
</tr>
<tr>
<td>25 - 28 Feb</td>
<td>146.5 ± 28.4</td>
<td>146.0 ± 3.1</td>
<td>31.4 ± 8.1</td>
</tr>
<tr>
<td>28 Feb - 3 Mar</td>
<td>102.5 ± 28.3</td>
<td>108.2 ± 5.8</td>
<td>23.0 ± 3.6</td>
</tr>
<tr>
<td>3 - 6 Mar</td>
<td>63.3 ± 10.0</td>
<td>63.0 ± 1.7</td>
<td>20.6 ± 0.8</td>
</tr>
</tbody>
</table>

**References**

