![NIVA]

Sampling of dissolved inorganic (DIP) and organic phosphorus (DOP) compounds in natural water by Diffusive Gradients in Thin-films (DGT)

Foto: Bjørn Faafeng

5 Phosphorus fractions in water TP, PP, TDP, DIP and DOP			
The Particulate P fraction	The Dissolved P fraction		
Total P fraction (TP)			
PP - Particulate P	Total Dissolved P (TDP)		
PP = TP – TDP	DOP = TDP - DIP		DIP
Particulate P	Dissolved Organic P		Dissolved Inorganic
Two phosphorus fractions in the DGT extracts, DOP and DIP			
Calculation of the two DGT fractions DGT–DIP and DGT–DOP			
DGT average concentration	$DGT\ Concentration\ =\ \ m\ /\ (t \cdot (DA/L)) \cdot$		
2 DGT fractions calculated	DGT-DOP (µg P/L)		DGT-DIP (µg P/L)

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REPORT

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Abstract

This report presents results from the work package 1 (WP1) of the EUTROPIA project, funded by the Research Council of Norway (RCN). The goal of WP1 was to improve the fractionation methods for phosphorus (P) compounds in water based on new auto-analytical techniques and passive samplers using Diffusive Gradient in Thin films (DGTs). Special focus was given to the Dissolved Inorganic and Dissolved Organic Phosphorus fractions (DIP and DOP) in water, as these contain the most bioavailable phosphorus compounds. The DGT adsorbents ferrihydrite (Fe-DGT) and the new titanium dioxide based (Metsorb™) adsorbent (Me-DGT) were compared. Both DGT adsorbents are known to collect the DIP fraction (orthophosphate). We wanted to study if the DOP fraction in water also could be collected by these adsorbents by using two low molecular weight model compounds adenosine monophosphate (AMP) and inositol hexakisphosphate (phytic acid, IP6). High collection efficiency and satisfactory extraction efficiency was achieved. A field study was performed in the Morsa-Vansjø catchment in stream water draining a forested, a mixed agriculture/forest and agricultural area. The results for the DGT-DIP fraction were comparable to the corresponding DIP fraction in stream water, while the DGT-DOP fractions showed some promising features. The results from the laboratory study and the field study is presented and discussed in the report.

4 keywords, Norwegian	4 keywords, English
1. DGT	1. DGT
2. Fosfor	2. Phosphorus
3. Eutrofiering	3. Eutrophication
4. Vann	4. Water

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Sampling of dissolved inorganic (DIP) and organic phosphorus (DOP) compounds in natural water by Diffusive Gradients in Thin-films (DGT)

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Preface

Oslo, 1. June 2014

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Contents

Summary

This report presents results from the Work Package 1 in the EUTROPIA project funded by The Research Council of Norway (RCN). The goal of WP1 was to improve the fractionation methods for phosphorus (P) compounds in water. Special focus was on the Dissolved Inorganic and Dissolved Organic Phosphorus fractions (DIP and DOP) in water, as these contain the most bioavailable phosphorus compounds. A new Continous Flow Analysis (CFA) auto-analytical technique was developed to determine the DIP and DOP fractions in water. The goal was furthermore to study the novel passive sampler Diffusive Gradient in Thin films (DGTs) for the collection of the DIP and DOP fractions. Two different DGT adsorbents were compared, the ferrihydrite (Fe-DGT) and the new titanium dioxide based (Metsorb™) adsorbent (Me-DGT). Both sampler types are known to collect the DIP fraction (orthophosphates) well. The uptake of the DOP fractions in water has not been previously studied by the DGT sampler. The two model compounds adenosine monophosphate (AMP) and phytic acid (inositol hexakis phosphate IP6) were used to examine the uptake of DOP compounds on the Me- and Fe-DGTs. Also a field study was conducted in tributary streams to the Lake Vansjø catchment.

Both the Fe- and Me-DGT samplers collected AMP and IP6 with high efficiency. The diffusion coefficient measured were 3.2 (AMP) and 1.3 (IP6) compared with 4.0 and $3.0 \cdot (10^{-6} \ cm^2/sec)$ derived from Buffle's equation. The deviations between measured and calculated values are caused by resistance of the diffusion membrane. The sampling precision obtained in the field study was 10-20% RSD. The accuracy was estimated to lie between 10 and 30%. The largest uncertainty is associated with the estimate of the diffusive boundary layer (DBL) and the diffusion coefficient (D). The adsorption capacities from 7 to 37 µg P permit 1 to 2 weeks sampling at concentrations below 50 µg P/L, without risk of overloading the adsorbents.

The DIP fraction consists mainly of orthophosphate species. The D value of these species decreases with increasing pH. A new correction function was developed for the orthophosphate species where the numerical value of D decreases with 0.15 per pH unit. The uncertainty of the D could be reduced to below 5% by this correction. Earlier a diffusion coefficient (D) of 6.0 $(10^{-6} \ cm^{-2}/sec)$ has been reported for the orthophosphate specie $H_2PO_4^-$. By comparing the concentration for the DGT-DIP fraction with the stream water DIP fractions, a D value of 5.5 was found at pHs between 6.5 and 7.5, which is in agreement with this pH correction function.

The DOP fraction consists of organic molecules with molecular weight (Mw) from 200 and up to at least 10000 Da. As the diffusion rates depend on Mw, one separate D value cannot be used for this whole Mw range. An equation developed by Buffle et al. (2007) is used to calculate the D value depending on Mw. Furthermore, the membrane resistance (R, i.e. the reduction of D in the membrane compared to the D in water) of some common DGT membranes was examined. The R factor increases with Mw, but more research is needed to understand the details of how the R term changes with Mw and other molecular properties of the organic compounds in the DOP fraction. By comparing the concentration for the DGT-DOP and the stream water DOP-fractions from agricultural soils, the D value was found to lie between 3.0 and 4.0 $(10^{-6} \ cm^{-2}/sec)$ at pH between 6.5 and 7.5. The best fit between DGT-DOP and stream water DOP was found at a relatively high D value of 3.7 $(10^{-6} \ cm^{-2}/sec)$. This indicates that run-off from agricultural soils consists of a DOP fraction dominated by low molecular weight compounds with Mw from 300 to 3000 Da.

The concepts for the operationally defined phosphorous fractions had to be revised in order to compare the DGT-phosphorus fraction with the corresponding phosphorus fractions in water. All the operationally defined phosphorous fractionation concepts for water had to be re-evaluated, harmonized with the recommendations in the Norwegian (NS) and international standard methods

(ISO), and further evaluated and updated according to a literature survey. This re-evaluation is described in a separate report by (Røyset et al. 2014).

The results from this first study on the use of DGTs for collection of dissolved inorganic and organic P fractions (DIP and DOP) in water show many promising properties. The field study performed in stream water draining a forested and agricultural area in the Morsa-Vansjø catchment, made it possible to display source patterns of the DIP/DOP fractions in the streams. In the water draining an agricultural field the DOP fraction constituted from 20 to 30% of total dissolved P. The DOP fraction was considerably higher (40- 60%) in runoff from a forest soil due to high content of dissolved organic material (DOM). The concentration of the DOP fraction was lower in the runoff from the forest soils (1 -5 µg P/L) than from the agricultural soils (5-20 µg P/L). However, since the forests cover 85% of the Morsa-Vansjø-catchment, the flux of organic P compounds (DOP) to Lake Vansjø is still considerable. The data for the different phosphorus fractions needs closer evaluation in order to quantify their contribution of the total P-flux to lake Vansjø.

DGTs represent a new valuable *in-situ* monitoring tool for determination of the dissolved DIP and DOP fractions in water, as the colloidal/particulate fraction is excluded by the pores size of the diffusion membrane (5-10 nm). Another useful feature is weekly or be-weekly averages where DGT display the nutrient status of these dissolved P compounds which may be relevant as threshold values to predict algae blooming.

The compounds in the DOP fraction have similar properties as the dissolved organic matter (DOM) in water. The membrane resistance data discussed in this report indicate that DGTs collect the low to medium molecular weight fraction (300 to 5000 Da) more efficiently than the high molecular weight fraction (5000 to 10000Da). The low molecular weight fraction of the DOM molecules is the best nutrient source for bacteria and algae in in water. If this assumption is correct, the DGT sampler will be a very useful tool to determine the most bioavailable fraction of the DOP molecules in water.

Sammendrag

Denne rapporten presenterer resultater fra WP1 i EUTROPIA-prosjektet finansiert av Norges forskningsråd (NFR). Målet med WP1 var å utvikle og forbedre fraksjoneringsmetoder for fosfor-forbindelser i vann, med spesiell fokus på de løste uorganiske (DIP) og organisk fosforfraksjoner (DOP), da disse inneholder de mest biotilgjengelige fosforforbindelsene. I mangel på gode norske begrep benyttes de engelske forkortelser DIP (Dissolved Inorganic Phosphorus), som for det meste består av ortofosfat-specier og DOP (Dissolved Organic Phosphorus), som er en heterogen gruppe løste molekyler som inneholder fosfor. En ny auto analytisk CFA teknikk (Continous Flow Analysis) ble utviklet for å bestemme DIP og DOP fraksjoner i vann. Målet var også å studere den passive prøvetakeren Diffusive Gradients in Thin Films (DGT) for oppfangning av DIP og DOP fraksjoner i vann. To forskjellige DGT adsorbenter ble sammenlignet: ferrihydrite (Fe-DGT) og den nye titandioksyd baserte (Metsorb ™) adsorbenten (Me-DGT). Begge adsorbenter er kjent for å samle opp ortofosfat specier. Opptak av DOP-fraksjonen i vann har ikke tidligere blitt studert med DGT prøvetakeren. De to modellforbindelsene adenosin monofosfat (AMP) og fytinsyre (inositol hexakisfosfat, IP6) ble anvendt for å undersøke opptaket av DOP forbindelser på Me- og Fe-DGTene. Det ble også gjennomført en feltstudie av DGT-ene i tilførselselver til innsjøen Vansjø.

Både Fe- og Me-DGT prøvetakere fanget opp AMP og IP6 kvantitativt. Diffusjonskoeffisienten (D) var 3.2 (AMP) og 1.3 (IP6) i forhold til beregnede verdier på 4.0 og 3.0 (10^{-6} cm^2/sec) utledet fra Buffles likning. Avvikene mellom målt og beregnet verdi er forårsaket av motstand i diffusjons-membran fra bl.a. negative ladning på molekylene. Prøvetakings presisjon oppnådd i felt studien var 10-20% RSD. Nøyaktigheten for DIP fraksjonen ble anslått til å ligge mellom 10 og 30%. Den største usikkerheten er forbundet med anslaget på diffusjons-grensesjiktet (diffusive boundary layer DBL) og D. Adsorpsjon-kapasitet på 7 til 37 µg P tillater 1 til 2 uker prøvetakingstid ved konsentrasjoner under 50 µg P/L uten risiko for metning av adsorbenten.

DIP fraksjonen i vann består hovedsakelig av ortofosfat-specier. D-verdien for de forskjellige ortofosfat-specier går ned med økende pH. En ny korreksjons-metode ble utviklet i dette arbeidet der den numeriske verdi av D reduseres med ca 0.15 per pH enhet. Tidligere D verdier for ortofosfat-speciet ($H_2PO_4^-$) har vært rapporter til ca 6.0 (10^{-6} cm^2/sec). Ved å sammenligne konsentrasjoner for DGT-DIP med feltmålinger av den tilsvarende DIP-fraksjon i vannprøver fra feltforsøkene, ble D funnet til å ligge på 5.5 (10^{-6} cm^2/sec) ved pH mellom 6.5 og 7.5 Nøyaktigheten for bestemmelse av DGT-DIP fraksjonen ble forbedret med denne pH korreksjon. Usikkerhet i estimatet for D for de aktuelle ortofosfat-specier ved den aktuelle pH i prøven kan reduseres til under 5%.

DOP fraksjonen består av en rekke organiske molekyler med molekylvekt (Mw) fra ca 200 til over 10000 Da. D for organiske molekyler avhenger av Mw, og man kan dermed ikke benytte bare en verdi som dekker hele dette området. Buffle et al. (2007) har utviklet en ligning som beskriver hvordan D endres med molekylvekt. Videre ble det studert hvordan membran motstanden (R, dvs. reduksjonen i D sammenlignet med D i vann) i de vanlige DGT membraner øker med Mw. Mer forskning er nødvendig for å bedre forståelse av for hvordan R varierer med endring av molekylvekt og andre molekylære egenskaper som ladning og form av molekyler som inngår i DOP fraksjonen. Ved å sammenligne konsentrasjoner for DGT-DOP med feltmålinger av den tilsvarende DOP-fraksjon i vannprøver fra feltforsøkene ble D for disse funnet til å ligge mellom 3.0 og 4.0 (10^{-6} cm^2/sec) ved den aktuelle pH mellom 6.5 og 7.5. Den relativt høye D indikerer at molekylvekten av DOP fraksjonen i vannprøver fra landbruksjord i dette området er lavmolekylære med Mw under 5000 og for en stor del under 1000 Da.

Konseptene for de operasjonelt definerte fosfor-fraksjoner oppsamlet med DGT måtte utvikles for å kunne sammenligne DGT-fosforfraksjon med de tilsvarende fraksjoner i vann. Også konseptene for

de tilsvarende operasjonelt definert fosforfraksjoner i vannet måtte revurderes, basert på anbefalinger i Norsk (NS) og internasjonale standardmetoder (ISO), og nærmere evaluert og oppdatert i henhold til en litteraturstudie. Denne re-evaluering er beskrevet i en egen rapport (Røyset et al. 2014).

Resultatene fra denne første studien på bruk av DGT for oppsamling av løste uorganiske og organiske P fraksjoner (DIP og DOP) i vann er så langt lovende. Feltstudien utført i bekkevann fra skogs- og jordbruks-areal i Morsa-Vansjø nedbørfeltet viste klare kilde-mønstre av DIP og DOP fraksjoner. I vann fra landbruksområdet utgjorde DOP fraksjonen 20 til 30 % av total løst P (TDP). DOP fraksjonen var betydelig høyere (40 - 60 %) i avrenning fra skogsjord på grunn av høyt innhold av løst organisk materiale (DOM). Konsentrasjonen av DOP fraksjon var lavere i avrenning fra skogsjord (1 -5 µg P/L) enn fra jordbruksjord (5-20 µP/L). Men siden skogen dekker 85 % av Morsa-Vansjø–nedbørsfeltet, er fluks av organiske fosfor forbindelser (DOP) til Vansjø fortsatt betydelig. Dataene for de ulike fosforfraksjoner fra EUTROPIA prosjekter trenger nærmere evaluering for å kvantifisere hvilke bidrag disse har for den totale fosfor fluks til Vansjø.

DGT representerer et nytt verdifullt *in situ* måle-verktøy for bestemmelse av DIP-fraksjoner i vann, da kolloider og partikler ekskluderes av de små porene på 5-10 nm i diffusjons-membranen. Dette betyr at DGT prøvetakeren måler ortofosfat-speciene som er de mest biotilgengelige formene av fosfor i DIP fraksjonen. En annen nyttig funksjon er mulighet til å måle ukentlige til to- ukentlige gjennomsnittsverdier. Slike DGT målinger av de løste ortofosfat-speciene kan være mer relevant for å forutsi algeoppblomstring.

En evaluering av oppfanging av løste organiske molekyler versus molekylvekt indikerer at DGT samler opp den lavmolekylære fraksjon (LMW) mer effektivt enn den høymolekylære (HMW) fraksjonen (større en 5000-10000 Da). LMW fraksjonen inneholder den delen av DOP molekylene som er den beste næringskilde for bakterier i vann. Dersom denne antakelsen er riktig, vil DGT være et svært nyttig verktøy for å måle de minste, lettest nedbrytbare og mest biotilgjengelige løste organiske fosfor molekylene i DOP fraksjonen i vann.

Abbreviations

D	The Diffusion coefficient D is normally expressed in the units 10^{-6} cm^2/sec for example by 3.0 10^{-6} cm^2/sec. During discussion in the text the D may be referred to by only the number without the exponent 10^{-6} (for example as the 3.0, meaning 3.0 10^{-6} cm)
Mw	Molecular weight expressed in Dalton (Da) or g mol^{-1}
DGT	The Diffusive Gradients in Thin Films sampler
Me-DGT	The DGT sampler with the Metsorb™ adsorbent
Fe-DGT	The DGT sampler with the Ferrihydrite adsorbent
MBM	Molybdate Blue Method
Ch.(Chapter)	If not written in full text, the abbreviation Ch. is used
Eq. (Equation)	If not written in full text, the abbreviation eq. is used
Buffle's equation	The relationship between Mw and D developed in the work by Buffle et al. (2007)
R	Membrane resistance factor (of a DGT membrane)
DOM	Dissolved Organic Material
DOC	Dissolved Organic Carbon
DNOM	Dissolved Natural Organic Matter (material)
NOM	Natural Organic Matter (Material)
LMW	Low Molecular Weight
HMW	High Molecular Weight
MMW	Medium Molecular Weight
FFF	Field Flow Fractionation

1. INTRODUCTION

1.1 Phosphorus - an important nutrient in water

Phosphorus (P), carbon (C) and nitrogen (N) are the most important nutrients in water. Phosphorus is present in the environment mainly at the P(V) oxidation state where the PO_4 group is ester-bound to organic or inorganic molecules ($RC\text{-}O\text{-}PO_3$). Knowledge of these forms is important for the understanding of the transformations, fate and effects of the phosphorus compounds in soils, soil water and the aquatic environment. Free inorganic orthophosphate ions are generally the main bioavailable form of phosphorus in water, and are rapidly assimilated. Decomposition and mineralization of complex phosphorus molecules to the small bioavailable orthophosphate ions is thus an important process as shown in Figure 1 (pathways for mineralization process). Phosphorus is often a limiting factor for growth in surface fresh waters. Large fluxes of phosphorus from sources such as municipal wastewater or in runoff from agricultural soils may thus induce eutrophication and blue-green algal blooming in receiving waters.

Figure 2 shows four fractions of phosphorus which can be separated by filtration and the digestion steps in the analytical chemical sample pre-treatment procedures used in laboratories. Figure 2 also indicate the bioavailability and susceptibility for mineralization/degradation to orthophosphate of each fraction. More details of the chemical compounds of phosphorus in each fraction are given in Table 1. While free orthophosphate species are immediately bioavailable, the release rate of orthophosphate groups from organic bound P compounds varies considerably depending on the physico-chemical characteristics of the organic material and the microbiological activity of soils and waters. The same is the case for the release rate of phosphorus from soil material or from suspended solids in water, which may vary considerably depending on the properties of the parent material.

The monitoring of bioavailable fractions of phosphorus compounds in water represents several analytical chemical challenges. Sensitive methods are required, as the free orthophosphate fraction is present at low concentration levels or even below the detection limit (typically 1 µg P/L for common methods). The largest analytical challenges are related to the separation of the fractions of total phosphorus and dissolved orthophosphate in samples with colloids and suspended particles. A recent work by Krogstad, Øygarden and Skarbøvik (2013) showed that several Norwegian commercial environmental analytical laboratories reported large deviations of suspended solids, total phosphorus and orthophosphate concentrations in synthetic water samples which were added suspensions of soil and sediment materials. Furthermore, the enhanced recognition of the role of dissolved organic phosphorus (DOP) compounds has raised the need for developments of s new phosphorus fractionation methods.

1.1.1 The EUTROPIA project

The EUTROPIA project studied the nutrient transport with special emphasis on phosphorus in the Morsa catchment to the eutrophic lake Vansjø. High phosphorus load is the major cause for the eutrophication of the lake Vansjø. Until now abatement actions enforced to reduce phosphorus loading have been focused on the anthropogenic sources from household wastewater and agricultural runoff. Despite numerous abatement actions to reduce these sources during the last 50 years, Vansjø still has occasional algal blooms in the summer season. Over the same time period the concentrations of DOC in streams draining the forested watersheds has doubled (Blankenberg et al., 2008; Skarbøvik, et al., 2012). The EUTROPIA project therefore questioned the role of phosphorus transport from the natural sources of the catchment (85% forested area), with a renewed interest on the DOP fraction in addition to the DIP and TP fractions.
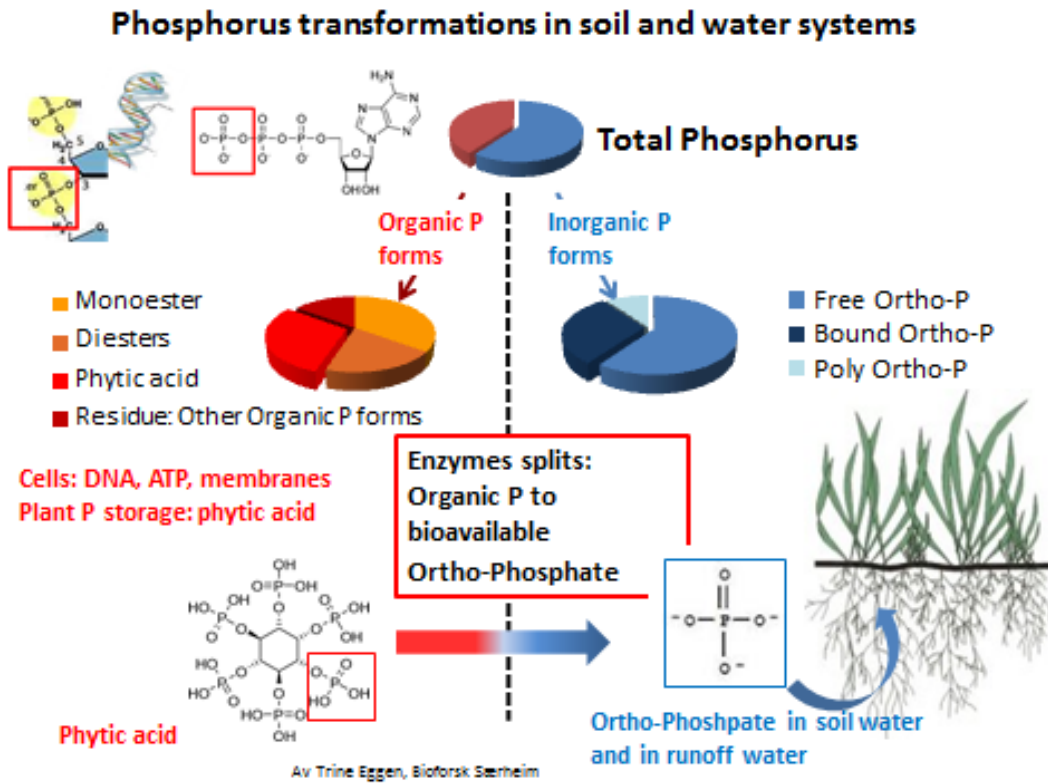
Figure 1.
Transformations and mineralization of phosphorus compounds from complex inorganic and organic molecules to free ortho-phosphate ions in soil, soil water and surface water (Figure by Eggen, 2013).

	Total Phosphorus (TP)			
	Total Particulate P (TPP)		**Total Dissolved P (TDP)**	
Filtration	Retained by a filter		Pass a filter	
Fractions	Particulate Organic P	Particulate Inorganic P	Dissolved Organic P	Dissolved Inorganic P
Denotation	**POP**	**PIP**	**DOP**	**DIP**
Compounds	Organic particulate matter, algae, bacteria, etc	Inorganic particulate matter, clays, hydroxides, etc	Phytines, Nucleotides,,P-sugars, P-lipids, humics	Free ortho-phosphate ions
Bioavailability	Low	Low	Medium	High
Mineralization to DIP	Slow	Slow	Medium	Instant

Figure 2.
Overview of the four phosphorus fractions found in natural water. The bioavailability and vulnerability to mineralization to orthophosphate species of the different fractions are indicated. The TP fraction is divided into the Total Dissolved and Particulate P fraction by filtration.

1.1.2 Developments of phosphorus fractionation methods

The WP1 of EUTROPIA addressed the need for development of new sampling and analysis methods for the determination of phosphorus fractions in water. These are needed in order to improve the understanding of the processes in the catchments for:
- Mobilization processes in the soils
- Transport through soil water flow-paths and in the tributary rivers and streams
- Surface water runoff from the forested and agricultural area of the Morsa-Vansjø catchment
- Internal cycling of phosphorous in the lake itself.

The study of DGT (Diffusive Gradients in thin Films) and auto-analytical methods were developed according to the original plan of WP1 of the project. This report describes the most important result for the developments of the DGT sampler. The need for the re-evaluation of the concepts (Chapter 2.1.5 below) was not planned in the project, but became more and more demanding during the last phase of the WP1 of the EUTROPIA project and after the projects was finished. This topic was clearly addressed by the main author during the RCN EUTROPIA Project Final Project Conference 30-31. May 2013 (Røyset, 2013a, 2013b). The fractionation concepts described in the standard methods were also compared with fractionation concepts commonly used based on a literature survey. This work is in progress in a separate report (Røyset et al. 2014).

1.1.3 Studies of the DGT sampler

The passive sampler Diffusive Gradients in Thin Films (DGT) was studied for the collection of Dissolved Inorganic P (DIP) and Dissolved Organic P (DOP) compounds in water by
- Examination of the properties of the phosphorus selective DGT adsorbents based on iron-hydroxide (ferrihydrite) and the new titanium dioxide based adsorbent with the brand name Metsorb™.
- Estimate DGT based diffusion coefficients (D) for compounds in the DIP and DOP fractions to be able to calculate the average DGT concentration for field deployments. For the DIP fraction the most important compound is the ortho-phosphate species. The DOP fraction contains a large number of compounds with molecular weight range from ca 200 to at least 10000Da. The estimations of appropriate D values were based on LMW model compounds, by the D vs molecular weight equation by Buffle, and further investigations of the resistance to free diffusion of these compounds groups in the DGT membrane.
- Establish new DGT fraction concepts needed to be able to compare the DGT fractions with the conventional DIP and DOP fractions in water.
- Compare the results achieved with the DGT-DIP and DGT-DOP fractions with the corresponding dissolved phosphorus fractions (water DIP and DOP) obtained by the new CFA methods (Chapter 2.1.4.).

1.1.4 Auto-analytical methods for efficient determination of phosphorus fraction in water

NIVA installed a new Continuous Flow Analyzer (CFA) in 2008 for the determination of up to seven fractions of carbon, nitrogen and phosphorus in water (see chapter 3.2). This CFA analyzer was used to develop new auto-analytical phosphorous fractionation methods for:
- The phosphorus fractions TP, TDP, DIP and DOP.
- Further improve these methods using water samples from the EUTROPIA project with demanding properties (TOC, particles etc).
- Improve the performance and efficiency of the system by assistance of NIVA's scientists (Blakseth and Hagebøe) in cooperation with the MSc student works by Parekh (2012) and Mohr (2010).

The performance of this CFA system is very promising. The results from this research are not reported here for space and time reasons as this needs separate reports.

1.1.5 Renew and harmonize the phosphorus fractionation concepts

During the EUTROPIA project water samples were analyzed at different laboratories, and datasets were compared from different monitoring program of the Morsa-Vansjø catchment (also delivered by different laboratories). Relatively large deviations were observed probably due to differences in the fractionation methods used. The different operationally defined fractionation methods and concepts derived from various versions of standard methods are used for both practical and historical reasons. It became clear that the operationally defined phosphorus fractionation concepts and methods in water had to be re-evaluated to:

- Evaluate the basis for the operationally defined fractionation concepts used for especially Total P (TP), Particulate P (PP), Total Dissolved P (TDP), Dissolved Inorganic P (DIP) and Dissolved Organic P (DOP).
- Improve the understanding of the groups of phosphorus compounds which are included in each of the fractions above, and how the fractions above can be separated in the operationally defined fractionation methods.
- Understand why different laboratories may obtain so large differences in what is expected to be the same fractions.
- Evaluate the fractionation concepts used in the Norwegian standard NS 4724/4725:1984, (valid 1984 to 2004)) and NS-EN-ISO 6878:2004 (valid from 2004), and in other ISO standards used by laboratories.

Based on the work above it was possible to develop a new approach to identify and compare all the operationally defined fractionation processes of different methods in a systematic way. This made it possible to examine which processes are the same and which differ, to decide if the fraction with the same denotation actually is expected to contain the same compound groups or not. I.e. are the results from the fractions delivered by different laboratories or by different methods comparable?

Based on this work new phosphorus fractionation concepts is proposed for harmonization of the operationally defined fractions delivered by different laboratories. This work is in progress in a separate report (Røyset et al. 2014).

1.2 Introduction to operationally defined phosphorus fractionation

The methods for fractionation of phosphorus compounds in water are based on **operationally defined fractionation procedures**. Several sample treatment operations, such as sampling, homogenization, resuspension, filtration, acid preservation, digestion and analysis are used to define the desired fraction. If these sample pre-treatments operations differ only in for example one of the steps, the results obtained for what is assumed to be same fraction, will not necessarily be comparable.

To understand the operationally defined fractionation procedures, the different sample pre-treatments steps must be clearly specified. A new set of concepts for the operational defined phosphorous fractionation methods in water has been developed. These are based on the recommendations in the Norwegian standard methods (NS-4724/4725:1984, NS-EN- ISO 6878:2004) and some additional ISO methods (see Røyset et al. (2014) for details about the relevant standard methods). These concepts have been further developed especially for the dissolved phosphorus fractions by reviewing the international literature in this area. The concepts are needed to be able to compare the phosphorus fractions obtained by the DGTs with the corresponding dissolved phosphorus fractions in water used in these standard methods. This is done in a separate work by Røyset et al. (2014). The main author therefore advice to read this report in

order to become familiar with the definitions of the operationally defined fractions obtained by different standard and/or laboratory methods

Figure 2 describes seven important phosphorus fractions in water, ie TP, PP, TDP, DIP, DOP, PIP and POP, which can be separated by common operationally defined phosphorus fractionation procedures. The PP fraction can be divided into PIP and POP by many operations (if required). In this work this separation into the PIP and POP fractions is not required as they are not collected by the DGT samplers. These fractions are mentioned in Figure2 for clarity but are not discussed in this work.

1.2.1 Determination of phosphorus fractions in water samples

When PIP and POP are removed, the seven fraction procedure is reduced to a five fraction procedure as described in Figure 3 (upper part). This five fraction procedure is very useful as it separates the three important dissolved phosphorus fractions TDP, DIP and DOP, while the PP fraction is left in one fraction. Figure 3 shows how these five fractions are obtained by the following operational processes:
- TP is first determined by PS (peroxodisulphate) digestion of a re-suspended, not filter, acid preserved sample.
- TP is divided into TDP and PP by filtration.
- TDP is obtained by PS digestion of this filtered acid preserved fraction.
- DIP is obtained by MBM analysis of the filtered acid preserved fraction. The dissolved organic compounds in the DOP fraction do not react with the MBM reagent (Moorleghem et al. (2013), and the DOP fraction can be obtained by difference. i.e DOP = TDP – DIP.
- PP can be obtained by difference between TP and TDP, or by the determination of the P fraction collected on the filter (digestion of filter). In this work PP is obtained by difference.

1.2.2 Determination of the phosphorus fractions in the DGT extracts

This five fraction procedure is the one needed for the comparisons with the DGT fractions. The TDP fraction in water is divided into the DIP and DOP fractions, which is required for comparison with the corresponding DGT-DIP and DGT-DOP fractions.

Briefly the procedure for the determination of the DIP and DOP fractions in the extracts of the DGT adsorbent is (see Chapter 3 for more details):
- The DGT adsorbents are first extracted to release the collected phosphorus compounds, acid preserved and the TDP and DIP fractions are determined (The TDP fraction is obtained after PS digestion of the extract and MBM analysis).
- DIP is obtained by MBM analysis of the preserved fraction. The dissolved organic compounds in the DOP fraction do not react with the MBM reagent. The DOP fractions can thus be obtained by difference. i.e DOP = TDP – DIP.
- The calculation from the amount of phosphorus in the DGT extract to the corresponding water concentrations is done by the DGT equation. The new DGT based phosphorus fractionation concepts DGT-DOP, DGT-DIP and DGT-TDP are used not to mix these with the input TDP, DIP and DOP fractions determined in the DGT extracts.
- The calculated phosphorus concentrations in (µg P/L) in water for the DGT-DIP and DGT-DOP fraction concentrations in (µg P/L) in water, depends on the diffusion coefficients (D) used. The values given in the Figure 3are "first choice" values if such D values have not been specially estimated for the purpose. The diffusion coefficients for the DIP and DOP fractions are very important to get accurate results. The estimation of the D is one of the most important topics of this work (see Chapter 2.4 and the discussion of these topics in Chapter 4.).

- The DGT-TDP concentration in Figure 3 is the sum of both DGT-DIP and DGT-DOP. It is important to notice that the DGT-TDP fraction is the sum of the calculated DGT-DIP and DGT-DOP concentration. It must not be mixed with the TDP of the DGT extract. DGT-TDP can be compared with the water TDP concentration when required.

Figure 3 is made to show that the DGT extracts are analysed in the same way as for the water samples. The section in the middle is shown to clearly state that the TDP; DIP and DOP in the DGT extract are determined as for the corresponding TDP, DIP and DOP fractions in water. The calculation of the DGT average concentrations are performed as shown in the lower part of Figure 3 (see Chapter 3 for more details).

Therefore Figure 4 is a simplified version of Figure 3 which hopefully is easier to quickly grasp and understand. The phosphorus in the extracts from the DGT adsorbent are analysed as if it was a normal water sample. The fractions TDP, DIP and DOP are achieved, and the corresponding DGT-DIP and DGT-DOP fraction are calculated as earlier explained.

5 Phosphorus fractions in water TP, PP, TDP, DIP and DOP		
The Particulate P fraction	**The Dissolved P fraction**	
Total P fraction (TP)		
Particulate P (PP)	Total Dissolved P (TDP)	
PP = TP – TDP	DOP = TDP - DIP	DIP
Particulate P	Dissolved Organic P	Dissolved Inorganic P
Three phosphorus factions in the DGT extracts, TDP, DIP, DOP		
Particles excluded by DGTs	Total Dissolved P (TDP)	
Fractions in DGT extracts	DOP = TDP – DIP	DIP
Calculation of DGT phosphorus fractions DGT-DIP, DGT-DOP and DGT-TDP		
DGT average concentration	$DGT\ Concentration\ =\ m\ /\ (t \cdot (DA/L)) \cdot$	
Diffusion coefficients (D)	DOP 3.0 10^{-6} cm^2/sec	DIP 6.0 10^{-6} cm^2/sec
2 DGT fractions calculated	DGT-DOP µg P/L	DGT-DIP, µg P/L
DGT-TDP- Sum fraction	DGT-TDP = DGT-DOP + DGT-DIP	

Figure 3.
Overview of the procedure for five phosphorus fractions in water (upper) and the two phosphorus fractions obtained by DGT sampler (lower). The concentrations of P in the DGT extracts are determined according to the procedures for TDP, DIP and DOP as for water (shown in the middle). The calculations of the DGT average concentration in water are done by the DGT equation as explained in chapter 3.

5 Phosphorus fractions in water TP, PP, TDP, DIP and DOP		
The Particulate P fraction	**The Dissolved P fraction**	
Total P fraction (TP)		
PP - Particulate P	Total Dissolved P (TDP)	
PP = TP – TDP	DOP = TDP - DIP	DIP
Particulate P	Dissolved Organic P	Dissolved Inorganic P
Two phosphorus fractions in the DGT extracts, DOP and DIP		
Calculation of the two DGT fractions DGT-DIP and DGT-DOP		
DGT average concentration	$DGT\ Concentration\ =\ m\ /\ (t \cdot (DA/L)) \cdot$	
2 DGT fractions calculated	DGT-DOP (µg P/L)	DGT-DIP (µg P/L)

Figure 4.
Overview of the procedure for the determination of five phosphorus fractions in water (upper) and the two phosphorus fractions obtained by DGT sampler (lower). The concentrations of P in the DGT extracts are determined according to the procedures for the dissolved fractions TDP, DIP and DOP as for water samples (upper part). The calculation of the DGT average concentration in water is done by the DGT equation as explained in Chapter 3.

1.3 Overview of P fractions and concepts

1.3.1 Overview of the most important phosphorus compounds in the different fractions

Table 1 shows the most important groups of phosphorus compounds and species which are included in the different operationally defined phosphorus fractions of Figure 2, 3 and 4. It is not possible to describe all the relevant phosphorus compound groups in water in full detail in this report. More information can be found in textbooks devoted to phosphorus in the environment such as Turner et al. (2004). Another good information source for this purpose is Wikipedia, by following the links given at http://en.wikipedia.org/wiki/Phosphoric_acids_and_phosphates.

Table 1.
Overview of the most important groups of phosphorus compounds and species found in the fractions described in Figure 2, 3 and 4. Filters with nominal pore size of 0.45 µm are recommended, but other nominal pore sizes are used in laboratories offering such fractionation methods. The phosphorus in each fraction is determined by the MBM method such as in NS-EN-ISO-6878:2004, but other instrumental methods may be used.

Fraction	Denotation	Phosphorus Compounds
Total P	TP	Total P contains all particulate and dissolved P compounds and species in the sample which can be digested to the ortho-phosphate species and determined by MBM. The digestion normally refers to the use of peroxodisulphate (PS), but other digestion methods may be used.
Total Dissolved P	TDP	All compounds in the DIP and DOP fractions passing a filter which can be digested (PS) to the ortho-phosphate species
Dissolved Inorganic P	DIP	Dissolved Inorganic P passing a filter: Ortho-phosphate (H_3PO_4, $H_2PO_4^-$ $\cdot HPO_4^{2-}$, PO_4^{3-}) and Non ortho-phosphate inorganic species (poly and meta phosphates)
Dissolved Organic P By difference	DOP = TDP-DIPP	Dissolved Organic P compound passing a filter, which can be digested by PS and included into the TDP fraction. The DOP fraction consists of a heterogeneous group of compounds with molecular weight (Mw) from ca 150 to ca 10000 Da, such as Phytic acid, DNA fragments, nucleotides (ATP, ADP, AMP), Phospholipids, Sugar phosphates, P-containing pesticides, P in Dissolved Organic Material (DOM), such as fulvic and humic acids. etc.
Particulate P By difference	PP =TP-TDP	When the PP fraction is expressed by difference, the PP fraction is expected to contain the phosphorus compounds associated to particulate and colloidal material retained by a filter. The PP fraction contains the phosphorus compound groups listed in the PIP and POP fractions below
Particulate P Digestion of filter	PP	The PP fraction collected on the filter is obtained by digestion. The PP fraction is expected to contain the same compound groups as for the PP fraction by difference as above, but this depends on the efficiency of the digestion process.
Particulate Inorganic P	PIP	Particulate Inorganic Phosphorus retained by a filter: Al, Fe, and Ca-phosphates, Phosphate adsorbed to clay minerals and Al/Fe hydroxides, and inorganic particulate and colloidal materials containing phosphorus
Particulate Organic P	POP	Particulate Organic Phosphorus retained by a filter such as algae, bacteria and organic particulate and colloidal materail containing phosphorus

1.3.2 Recommended concepts

Table 2 summarizes the concepts recommended by Røyset et al. (2014) both by the conventional phosphorus fractionation methods in water and by the use of DGT sampler. The concepts used for the DGT are harmonized with the corresponding DIP and DOP fractions for water. The large number of fractionation concepts used in the literature are discussed and evaluated by Røyset et al. (2014). Table 1 gives overview of the most common abbreviations used in this report. Table 2 gives overview of what the main author recommends as concepts for the future.

Table 2.
Abbreviations and concepts used for the determination of phosphorus fractions in water recommended by Røyset et al. (2014).

	Total and particulate phosphorus fractions
TP	Total Phosphorus in PS (Potassium Peroxodisulphate) digested water samples
PP	Particulate Phosphorus. Determined as the difference between TP and TDP. PP may also be determined by digestion of particles retained on the filter to transform the phosphorus to the orthophosphate species
PIP and POP	Particulate Inorganic and Organic Phosphorus collected on a filter. Several digestion/extraction procedures are available for the determination of the separate PIP and POP fractions.
	Dissolved Fractions
TDP	Total Dissolved Phosphorus in filtered samples after PS digestion
DIP	Dissolved Inorganic Phosphorus after filtration, but without PS digestion
DOP	Dissolved Organic Phosphorus as difference between TDP and DIP, i.e. DOP = TDP - DIP
	Fractions only related to the DGT sampler
DGT-DIP	Dissolved Inorganic Phosphorus determined by the DGT sampler (Determined as the DIP fraction in the DGT extracts by MBM without PS digestion and then calculated to the DGT-DIP concentration)
DGT-DOP	Dissolved Organic Phosphorus determined by the DGT sampler (Difference between the TDP and DIP fraction in the DGT extract and then calculated to the DGT-DOP concentration)
DGT-TDP	Total Dissolved Phosphorus by the DGT sampler calculated as the sum of the DGT-DIP and DGT-DOP fractions above.

1.4 Sampling of phosphorus compounds in water by DGTs

The passive sampler Diffusive Gradient in Thin films (DGT) has since the introduction by Davison and Zhang (1994) become a very useful sampling tool for dissolved ions and molecules in water. DGTs collect ions on an adsorbent membrane placed behind a diffusion membrane. The first DGT sampler developed was equipped with a chelating imino-diacetate ion exchange resin (Chelex™) as adsorbent and collected di- and tri-valent metal ions. Up to about 30 metal ions could be collected as demonstrated in the first 55 multi element DGT map of the Chelex™ based DGT (Garmo, Røyset et al., 2003). DGTs accumulate ions linearly over time and produce thereby an average concentration. Since DGTs accumulate ions during the deployment period, it is possible to determine very low concentration of the analyte. This pre-concentration is especially useful for the determination of free orthophosphate species in water when the P-concentration is reduced to low ppb levels by algal grazing.

Originally the DGT was developed for the collection of inorganic metal cations in water. Several new adsorbents have the last years broadened the application area of the DGT sampler from metal cations to also include several anions and oxyanions such as orthophosphate. For this purpose positively charged adsorbents had to be developed. The first adsorbent for orthophosphate was based on iron hydroxide (called Ferrihydrite DGT, Fe-DGT), as studied by Zhang et al. (1998). Røyset, Bjerke, Eich-Greatorex, Sogn, Almås, (2004) and Sogn, Eich-Greatorex, Røyset, Øgaard,

Almås (2008) studied the Fe-DGT for collection of phosphate, arsenate and selenite. They found that the Fe-DGT collected these oxyanions with high efficiency. Lately the DGT sampler has also shown considerable promise for the collection of low molecular weight dissolved organic molecules such as pesticides and antibiotics (Chen, Zhang and Jones (2012).

Recently a titanium dioxide (TiO_2) based adsorbent (called Me-DGT based on the brand name Metsorb™) has shown to be promising. The Me-DGT was reported to have higher adsorption capacity than ferrihydrite. Applications of the Fe and Me-DGTs have been studied by several Australian groups (Panther et al., 2010, 2011, Mason et al., 2005, 2008, 2010). Lately the application of Metsorb™ has been expanded to several other negatively charged oxyanions (Panther et al., 2013, Price, Teasdale and Jolley 2013). The latter concluded that ferrihydrite may have considerably higher adsorption capacity than earlier expected compared to the Metsorb™ adsorbent.

1.4.1 Objective of the study of the DGTs in this is work

The strong affinity of Fe-DGT and Me-DGT for phosphate ions makes them also potentially suitable adsorbents for dissolved organic phosphorus (DOP) compounds in water. When this project was planned in 2008, the use of DGTs for the collection of dissolved organic phosphorus compounds in water had not been reported. The objective of this work was therefore to study the uptake of low molecular weight DOP compounds in addition to orthophosphate species on the two DGT adsorbent ferrihydrite and Metsorb™. The Fe-DGT and Me-DGTs were therefore studied for these compounds in the laboratory and later in the field for simultaneous collection of the DIP and DOP fractions in surface water.

Laboratory study
An initial laboratory study of the Fe-DGT sampler was performed by the MSc work by Mohr (2010). Later this work was broadened by a laboratory study of both the Fe- and Me-DGT samplers by the MSc work of Parekh (2012). The goal of the laboratory study was to determine the diffusion coefficients (D) for two model compounds simulating low molecular weight DOP compounds in water: I.e. adenosine monophosphate (AMP) and phytic acid (inositol-hexakis-phosphate-IP6). The diffusion coefficients (D) achieved from the model compounds was used to develop understanding on how to estimate D values for similar LMW organic phosphorus compound collected by the DGT sampler in the field.

Field study
The Fe- and Me-DGT samplers were used to collect DIP and DOP compounds in surface water runoff from the MORSA-Vansjø catchment in south eastern Norway, as described in the MSc work by Parekh (2012) and Mohr (2010). The Fe- and Me-DGT samplers were also investigated to compare the performance for the collection of the DOP and DIP compounds. The data for the DGT fractions were then compared to the same fractions in water to investigate if the diffusion coefficients found in the laboratory study could be used for field sampling, or if these had to be modified for this purpose.

Clarification of concepts for the P fractions achieved with DGTs
During the work the concepts and procedures used for the new phosphorous fractions achieved by the DGTs and the conventional operational phosphorus fractionation methods for water had to be clarified. It turned out necessary also to re-evaluate the operationally defined phosphorus fractions in water based on the NS 4724/4725:1984 and NS EN ISO 6878:2004 methods and compare these with those used in the literature. The new phosphorus fractionation procedures are shown in Figure 2, 3 and 4. The compounds expected to be found in each fraction is outlined Table 1. The proposed new concepts are shown in Table 2. The background for these new concepts is also further discussed in a separate report by Røyset et al. (2014).

This was necessary in order to be able to compare the DGT fractions with the corresponding fractions in water. The DGT-DIP and DGT-DOP fractions have not the same operational definitions as for the corresponding DIP and DOP fractions in water. The DOP fraction in water obtained by the conventional fractionation methods depends on the cut-off for the filter (ca 0.45 µm for the most common filters used). 0.45 µm correspond to a fraction of large macromolecules much larger than those which can pass a DGT membrane with pore size of 5-10 nm. The sampling and preservation regime for grab samples of water are also different from DGTs.

The average DGT concentration over the deployment period, depends on the diffusion coefficient, and the factors such as Mw cut-off depending on the pore size of the DGT membrane, in addition to other membrane factors. The pore-size of the most common agarose polyacrylamide (APA) DGT membrane is 5 -10 nm (0.005 to 0.01 µm). The relationship between molecular weight, molecular diameter and D is shown in Table 11 and further discussed in chapter 4.3. A molecular diameter of 5 and 10 nm correspond to molecular weights of ca 50 000 and 500 000 Da respectively (Table 11). Thus a DGT membrane with a pore-size of 5 nm should have a molecular size cut-off around 50000 Da.

Nevertheless, the intention was to compare the DGT fraction with the corresponding stream water grab samples fractions: DGT-DIP with water DIP, DGT-DOP with water DOP and DGT-TDP with water TDP. Earlier works have shown that DGTs deliver results which are comparable to water grab samples for the DIP fraction. One of the intentions of this work was to examine if the same was the case for the DOP fraction.

1.4.2 Phosphorus compounds collected by DGTs

The DGT adsorbents collect ions or molecules which pass the membrane of the DGT. Below a molecular size cut-off of 5 nm (ca 50000 Da for the APA membrane) the DGT is expected to collect the following phosphorus compound groups in water:

- The DIP fraction
    a. The orthophosphate species and possibly polyphosphates. There is little documentation of the adsorption efficiency of polyphosphates by DGTs, but they are presumably adsorbed with the same efficiency as the orthophosphate species. However, polyphosphates are usually present in only minor amounts in natural water compared to ortho-phosphate species. Therefore the general assumption is that the DIP fraction collected by DGTs in natural water will mostly consist of orthophosphate species.
- The DOP fraction in water consists of several compound groups as shown in Table 2.
    a. Low molecular weight dissolved organic phosphorus compounds with Mw below 1000 Da such as phytins, nucleotides, sugars etc. These are considered to have defined chemical structures and molecular weight, and can be identified by separation techniques such as HPLC.
    b. DOM compounds containing phosphorus in water which can pass the pores of the membrane, such as fulvic acids humic acids. Table 3 shows how these are classified according to molecular weight.
    c. Other DOM compounds in water containing phosphorus which can pass the DGT membrane. The normal APA membrane issued in this work, but it should be noted that other membranes with larger pores can be used, such as agarose membranes

The following compounds are not expected to be collected by the DGTs
- Colloids with diameter above 5 to 10 nm are excluded by size
- Colloids less than 5-10 nm diameter has slower diffusion rates and may also partly be excluded by charge and membrane resistance, as well as not being captured by the adsorbent.

1.4.3 Content of P and properties in DOM materials in water

The natural organic material (NOM) dissolved in water may be classified as Dissolved Organic Matter (DOM) or Dissolved Natural Organic Material (DNOM). For simplicity we use the concept DOM in this report as this is a more general concept. The DOP fraction is considered as a part of the DOM fraction, as DOM molecules containing phosphorus. The collection efficiency of the DGT adsorbents for the different dissolved organic compound may vary. Limited information is available about the diffusion coefficients in the DGT membrane of molecules in the DOM and DOP fraction in the Mw range 200 to 10000 Da. This is probably the largest challenge by the use of DGTs for the collection of the DOP fractions. More information about the properties of the DOP compounds in water is given below. The diffusive properties of the DOM/DOP compounds are discussed in in 4.3 and the elemental composition such as C:P ratios is discussed in chapter 4.7 .

Despite that DOM is the main natural transport mechanism of the micro nutrient phosphorous from terrestrial to aquatic environment, the knowledge regarding the content of P in the dissolved natural organic matter is limited. The Redfield atomic ratio 106:16:1 of C:N:P, found in phytoplankton and throughout the deep ocean, are commonly also applied for aquatic DOM (Perdue, 2009, Spivakov et al. 1999). In soil organic matter Cleveland and Liptzin (2007) reported a surprisingly constrained mean C:P molar ratio in soils of 186:1. Kortelainen et al. (2004), reviewing articles on allochthonous DOM in European catchments as a part of the DOMAINE project, found a large span in DOC and DOP concentrations, ranging between 40 to 4 000 and 0.05 to 2 µM, respectively. Within this project, studying 34 watersheds in France, Denmark, Wales, and Finland, they found C:P ratios of 326, 545, 780 and 4400, respectively. In a study of the DOM in the Everglades, Florida USA, Ged and Boyer (2013) found that the DOP was not correlated with the DOC. Furthermore, they found that the high molecular weight fraction (i.e. > 10 kDa), accounting for 40% of the DOP, had C:P ratios of 1 037 and 2 200 during spring and summer sampling campaigns, respectively. This is close to the molar ratios found for the whole water of 1 050 and 1 675. Most of the DOP (~45%) were found in the size fraction < 1 kDa, with a C:P ratio of 285 and 810. This they explain by that large uncharged DOP molecules pass through pores smaller than their specified molecular weights while small negatively charged DIP molecules do not pass through the ultra-filtration membrane. Consequently there is an inherent interaction between the membrane and DIP in the water matrix yielding lower DIP levels and higher DOP levels in the < 1 kDa size range. The three middle molecular weight fractions (1-3 kDa, 3-5 kDa and 5-10 kDa) were the most P deficient, with ratios 5 – 10 times greater than that of the whole water. A survey of concentrations of organic C, N, and P, elemental ratios in DOM of the Everglades (Qualls and Richardson, 2003) report C:P ratios ranging from 1 000 to 13 000. Based on these few studies it is apparent that the C:P ratio of aqueous DOM varies considerably, but is clearly higher than the Redfield ratio by about one order of magnitude.

1.4.4 Molecular weight distribution of DOM and DOP compounds in water

Molecular weight is one of the most used parameter to estimate diffusion coefficients of macromolecules like DOM and DOP in water (see chapter 4.3). The literature contains few data on the diffusion coefficients of typical dissolved organic phosphorus compounds in water. The best source is to use data from DOM compounds in water, as the molecules of the DOP fraction have similar properties as DOM. However, to use the data from the literature, we need to understand how the DOM materials are classified with respect to molecular weight and other relevant properties.

The DOM group may be classified according to several criteria: By molecular weight, by its elemental composition (C, H, O, N, S), by basic molecular carbon structures (aliphatic, aromatics, sugars, lipids etc), by the functional groups, by its charge properties (based on how the functional groups protonize), etc. Perdue and Ritchie (2003) has a comprehensive review of

these properties which also is updated by Perdue (2009). The most important Mw classes obtained from these reviews are summarized in Table 3.

These classifications criteria are fairly coarse, but are needed for collection and evaluation of diffusion coefficients from the literature for DOM compounds. In many works the properties are reported for DOM molecules classified as fulvic of humic acids (Lead et al. 2003, Buffle et al. 2007), or based on molecular weight ranges of the DOM molecules (LMW, MMW, HMW) obtained by ultrafiltration (Ged and Boyer (2013)). The evaluations needed to estimate the diffusion coefficients of the DOM and DOP molecules in water are discussed in chapter 4.3.

Table 3

Classification of DOM compounds in water as belonging to humic substance (HS) groups, or just by molecular weight (Mw). The Mw ranges are proposed by co-author Vogt based on the reviews by Perdue (2009) and Perdue and Ritchie (2003). The upper Mw ranges are somewhat arbitrary, and may differ from other authors, so these are therefore indications of the most common Mw ranges encountered in water.

Compounds	Notation	Mw range
Dissolved Organic Matter	DOM	200 – 10000 Da
Natural Organic Matter	NOM	200 – 10000 Da
Fulvic acids	FA	700 to ca 3000 Da
Humic acids	HA	2 000 to 6600 Da.
Humic substances	HS	700 –at least 10000 Da
Low molecular weight	LMW	<1000 Da
High molecular weight	HMW	1000 to 10000 Da
Large humic molecules		>10000 Da
Molecular weight ranges	Molecular Weight range obtained by f. ex. Ultra-filtration	<1000 Da 1000 – 3000 Da 3000 – 5000 Da 5000 – 10000 Da >10000 Da

1.4.5 DGT as a tool to determine the bioavailable phosphorus fractions

One of the most interesting applications of the DGT sampler is as a tool to determine the bioavailable fractions in water, which can be used to predict the toxicity of metal ions. Also deficiency can be predicted both for metal ions and for nutrient ions such as phosphorus. This is described in the basic review by Degryse, Smolders, Zhang & Davison (2009). Several promising application have been published (Zhang, Mason et al (2013), Six, Smolders et al. (2012); Tandy, Mundus et al. (2011)). The possibility to separate DOP and DIP fractions of P will be of great additional value in this application area. Thus, DGTs are expected to collect the dissolved inorganic ortho-phosphate species and a dissolved organic phosphorus fraction in water with molecular weight from ca 200 up to ca 10000 Da. These groups are believed to contain the most bioavailable (DIP) or potentially bioavailable phosphorus fractions (DOP) in water. This makes DGTs an attractive tool for selective collection and determination of the most bioavailable phosphorus fractions in water.

1.5 The Morsa-Vansjø catchment

The work was conducted as part of the interdisciplinary EUTROPIA project, funded by RCN from 2008-2012. Lake Vansjø is an eutrophic lake in the Morsa catchment, located in south eastern Norway as shown in Figure 5. Further details of the catchment are given by Skarbøvik and Bechmann (2010), Skarbøvik, and Bechmann (2013), and Skarbøvik and Haande (2012).

The Morsa catchment consists of a predominantly forested area in the north and west, and a more agricultural dominated area in the south-east. Lake Vansjø receives inputs from one large and several smaller streams. The Lake Vansjø has several conflicting user interests: Drinking water supply for Moss city, water supply for irrigation to farmers, local sport fishery, water sports, bathing, leisure, holiday cottages, and important habitat for local flora and fauna etc.

The lake receives a high nutrient load from natural P rich marine soils and the anthropogenic activities in the catchment. This has led to frequent annual algal blooms in the summer season during the last 50 years. The Vansjø catchment has undergone numerous abatement actions (Orderud & Vogt, 2013), to reduce the nutrient loads such as, 1) Renovation of the local municipal sewage system to practically exclude all household waste water input. 2) Several actions to reduce agricultural runoff: Reduced P fertilizer use, changed agricultural practices such as reduced autumn ploughing especially in high erosion zones, landscape forming actions (collection dams, grass belts) to reduce and collect surface erosion. The municipalities of the whole Morsa catchment have developed a cooperation organization where these parties work together to assess and further develop abatements actions (the MORSA cooperation group).

The success of these abatement actions to reduce the eutrophication of the lake has been less than expected. Local Municipality authorities, local stakeholder, NGOs, regulators and environmental advisors still have gaps in the understanding of why the abatements have not improved the water quality satisfactory. Modelling studies within the EUTROPIA project indicates still lack of data for some major sources and processes for nutrient loads from the catchment to the lake (such as DOP compounds).

The EUTROPIA project had a strong focus on the role of Dissolved Organic Material (DOM) in mobility and transport processes governing the flux of P to the surface water in the form Dissolved Organic Phosphorus (DOP) compounds. In runoff water from forest and agricultural soils we may expect that the molecular weight ranges (Mw) of the LMW compounds and other compounds in the DOP fraction are from about 200 to at least 10 000 Da.

In addition to the P loading from the agricultural areas, the forested areas, constituting 85% of the catchment area, and have a significant contribution to the total P loading to the lake. The lack of adequate knowledge of the amount of P in DOM in the forest runoff water limits sound estimates. The flux of P from the forested areas has been estimated to account for approximately 40% of the total P loading to the lake (Parekh 2012; Vogt, 2012). The concentration of DOM in forest runoff has almost doubled over the last 20 years (Blankenberg et al., 2008). Particular focus has therefore been brought onto the fraction of LMW organic phosphorus species. Another knowledge gap is how easily the DOP fraction degrade and transform into a bioavailable P fraction (DIP) which algae subsequently can use as P-nutrient source.
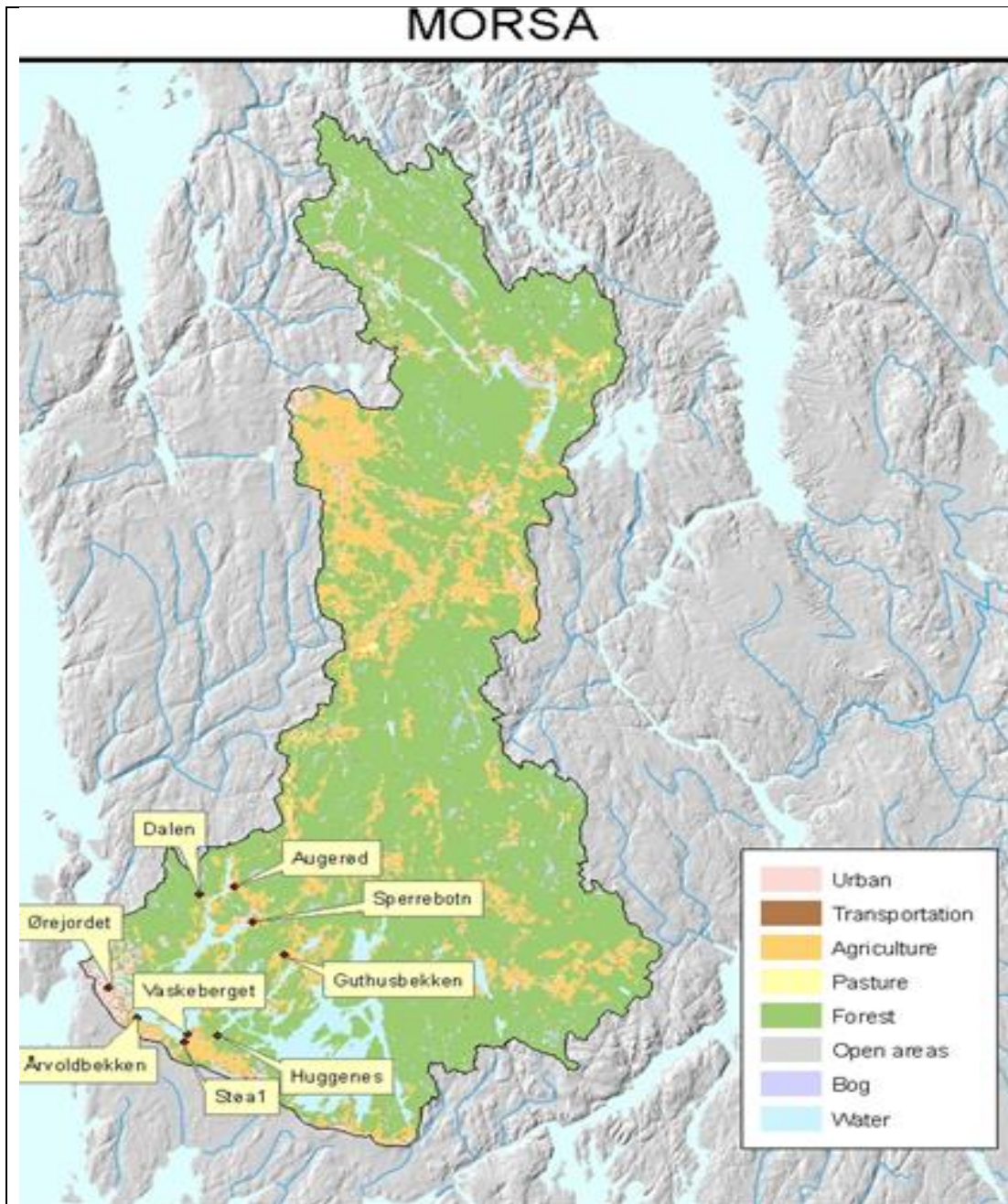
Figure 5.
The Morsa catchment with Lake Vansjø and predominant landscape types (Norwegian Forest and Landscape Institute (2011), revised by Alexander Engebretsen, and as shown by Parekh (2012). Sampling positions of the EUTROPIA project are shown: Dalen (forest) up to the left, Støa (agriculture) and Huggenes (mixed agriculture and forest) down to the right.

2. MATERIAL AND METHODS

2.1 Chemicals

The test compounds used as models for DOP compounds in water, were Adenosine monophosphate (AMP, brand Merck, purity >99%) and Inositol hexakis-phosphate (IP6, Brand Sigma, purity >95 %). See Figure 6 for details on chemical structure and properties. All chemicals used (NaCl, Na- and NH_4COOH, NaOH etc.) were of pro analysis (p.a.) quality or similar (see Parekh (2012) for details). The chemicals used for determination of phosphorus by the SKALAR SAN CFA analyser are described in Chapter 3.5.

The DGTs were purchased from DGT research Ltd (www.dgtresearch.com). The ferrihydrite DGTs are the type offered from DGT Research Ltd. The Metsorb™ HRMP material is a finely ground hydrated TiO_2 powder with particle size < 50 µm and is the same as applied by Panther et al. (2010). This was purchased from Graver Technologies, US, ((http://www.gravertech.com/pr_ads_metsorb_HMRP.html). It was cast into DGT adsorbent membranes of 0.4 mm thickness as described by Panther et al. (2010) by DGT Research Ltd. This is the same casting procedure as used for the Ferrihydrite adsorbents.

Both thee Metsorb™ and Ferrihydrite DGTs were packed with the normal open pore diffusion membranes and protection filter in the same way as for normal DGTs. This membrane is the patented membrane by DGT Research Ltd and denoted open pore APA gel (agarose polyacrylamide) with a pore size of 5 -10 nm.

Ortho-phosphoric/-phosphate species		
Specie	pK_a	
H_3PO_4	2.12	
$H_2PO_4^-$	7.21	
HPO_4^{2-}	12.67	
PO_4^{3-}		

AMP - Adenosine mono phosphate $C_{10}H_{14}N_5O_7P$, Mw 347.22 g/mol AMP has 3 pK_a values: 0.9, 3.8, 6.1	

IP6, Inositol hexakis-phosphate (phytic acid) IUPAC name: 1r,2R,3S,4s,5R,6S)-cyclohexane-1,2,3,4,5,6-hexayl hexakis[dihydrogen (phosphate)] $C_6H_{18}O_{24}P_6$, Mw 736 g/mol IP6 has 12 pK_a values ranging from 1 to ca 12 1.1, 1.5, 1.5, 1.7, 2.1, 5.7, 6.85, 7.6, 10, 10, 12 and 12, (.Costello et al. (1996))	

Figure 6.
Chemical structures and properties of the orthophosphate species, AMP and IP6 used in this study.

2.2 Phosphorus analysis by the CFA analyzer

2.2.1 Description and principles of the CFA analyser

The determination of phosphorus in the different fractions in the water samples and in the DGT extracts, were performed using an air segmented Continuous Flow Analyser (denoted CFA, also often air segmented CFA). This analyser is a SKALAR SAN analyser (delivered by SKALAR, see http://www.skalar.com/analyzers/automated-wet-chemistry-analyzers). The analyser is customized by the requirements specified by NIVAs laboratory, and is under development at NIVA. It is beyond the scope of this report to explain the details on how this CFA analyser works. The general methods and principles are explained Table 4.

Table 4.
Flow scheme for the SKALAR CFA analyser consisting of 7 channels divided into two units run in parallel. One unit is set up to run the dissolved nutrients and the other one for the total components. In this customized system the TOC and TOT-N channel use the same PS digestion manifold. The output of this combined PS manifold is then divided into two different detection channels/chemistries. The temperature of 107 ^{o}C is achieved with a backpressure unit of 1.4 bar.

UNIT	Ch.	Parameter	Method principle	Detection	Detection
THE NUTRIENT UNIT	1	PO4-P	Acid MBM chemistry	Colorimetric	880 nm
	2	NO3-N	Cd/Griess reaction	Colorimetric	520 nm
	3	NH4-N	Ortho-phthaldialdehyde	Fluorescense	425 nm
	4	Si	Neutral MBM chemistry	Colorimetric	880 nm
			On line PS Digestion	**Detection**	
THE TOTAL UNIT	5	TOT-P	Acid PS pH ca 1, 107°C, 1.4 bar	MBM chemistry Colorimetric	880 nm
	6	TOT-N	Alkaline PS pH 10, 107°C 1.4 bar	Cd/Griess reaction Colorimetric	520 nm
	7	TOC		CO_2 stripping IR detection	1500 nm

2.2.2 CFA methods CFA methods

The chemicals and reagents used for determination of phosphorus fractions (according to Table 4) are described in the methods provided by SKALAR. These methods are confidential. These methods are customized for use at NIVA (such as NIVA method D10), and integrated into NIVAs Quality Assurance system, where the details are described. These methods are also confidential. Here only the principles of the methods are explained.

The CFA procedure for determination of P in the samples is based on the ISO standard method (15681-2: 2003) for the determination of orthophosphate (PO4-P) and total phosphorus (TOT-P). The total and total dissolved fractions are achieved through on-line peroxodisulphate oxidation (PS) in the CFA system. This PS oxidation manifold is set up to decompose the non-ortho-phosphate species. The detection of the released orthophosphate species from the PS oxidation is done by the conventional MBM-chemistry in the CFA manifold. The digestion efficiency of the PS system is monitored by Quality Control Digestion Efficiency (QCDE) samples. For the TOT-P channel AMP is used. A digestion efficiency of 90% or better for AMP is achieved both for the manual digestion NIVA method and in the CFA system (see Røyset et al. (2014) for more details). Most of the organic molecules of the DOP fraction d extracted from the DGTs are expected to be fairly easy to digest by the PS reagent of this CFA analyser system. When the digestion efficiency AMP is above 90%, the same high digestion efficiency is expected also for DOP compounds in the

TPD fraction. More details about the digestion efficiency of AMP of other organic compounds are discussed by Røyset et al (2014).

2.2.3 Denotation of parameters delivered by the CFA analyser

Both the TOTAL and NUTRIENT units in Table 4 are run in parallel to determine both Total phosphorus (TOT-P) and orthophosphate (PO4-P) in the same run. The CFA instrumental method has its own denotation for the parameters delivered. Therefore the denotations in Table 5 are only related to the CFA analyser methods. The interpretation of the parameters delivered by the CFA analyser (for example PO4-P and TOT-P) must therefore be related to the sampling system. For example TP in water is achieved by the TOT-P channel of the SKALAR TOT-P method. The DIP fraction in water is achieved by the Channel for PO4-P by the SKALAR PO4-P method. These details are described in the laboratory quality system and in the individual methods. It is beyond the scope of this report to explain all this in detail here. As explained by Røyset et al (2014) a number of old concepts are inherited both from the NS/ISO standards methods, from the literature and from former laboratory procedures and practices.

2.2.4 Sample collection, preservation, filtration and matrix matching of standards

Collection and storage: Water samples are collected in PE plastic bottles, and stored dark and cool (4^oC). Before preparation of subsamples, the original PE bottles were shaken to resuspend all particles to collect representative subsamples. All the subsamples above were, unless otherwise stated, prepared in the 30 ml PP tubes used for the autosampler of the CFA system (Sarstedt PP centrifuge tubes with PE caps). All tubes were acid cleaned by sulphuric acid before use.

Preservation. All samples and sub-samples for phosphorus fractions (and nutrients in general) were preserved to 0.08 N sulphuric acid (1.0 mL of 8 N sulphuric acid added to 100 mL sample). After preservation the samples were stored at least over night, but could also be stored for longer when required. This 0.08 N preservation is the general purpose preservation of all water samples at NIVAs laboratory for the determination of all C, N and P nutrients. This preservation is also recommended by the Norwegian Standard (NS 4724-4725:1984) for the determination phosphorus fractions in water. My opinion (the main author) is that the 0.08 N preservation is both more efficient and ensures safer storage of samples, than the so-called neutralization recommended in NS-EN-ISO 6878:2004 (see Røyset et al. (2014)) for further discussions on the preservation methods and general evaluation of the standard methods).

Matrix matching: The 0.08 N sulphuric acid is therefore the general matrix used for all the phosphorus and nutrient analysis at NIVA's laboratory. All calibration standards are also preserved in this 0.08 N sulphuric acid matrix.

Filtered sample: Depending on the purpose, different filter types were used in this project: Conventional membrane filter with nominal pore size of 0.45 µm, Whatman GF/F (pore size 0.7 µm) or GF/C (pore size 1.2µm) glass microfiber filters. Filtered subsamples were preserved to 0.08 N sulphuric acid. The P in the preserved TDP and DIP fractions were determined by the CFA analyser.

Unfiltered sample. Re-suspended subsample were collected and preserved to 0.08 N sulphuric acid. Then the TP – (and the unfiltered NFAP fraction if required), were determined by the CFA analyser or by the manual procedure for TP at NIVA.

2.3 Laboratory study of the DGT

2.3.1 The DGT exposure chamber

The DGT exposure chamber developed by Garmo, Røyset, et al. (2003), was used for the laboratory study of uptake of AMP and IP6 on the DGTs. As shown in Figure 7, this consisted of a 50 L container with a rotor holding 3 central rotating disks. These laboratory experiments were conducted as part of the MSc works by Mohr (2010) and by Parekh (2012).

50 L exposure tank (A). DGT mounting rotor (B)	DGT sampler
	Some DGTs can be seen mounted in the upper rotor plate in the picture B (middle)
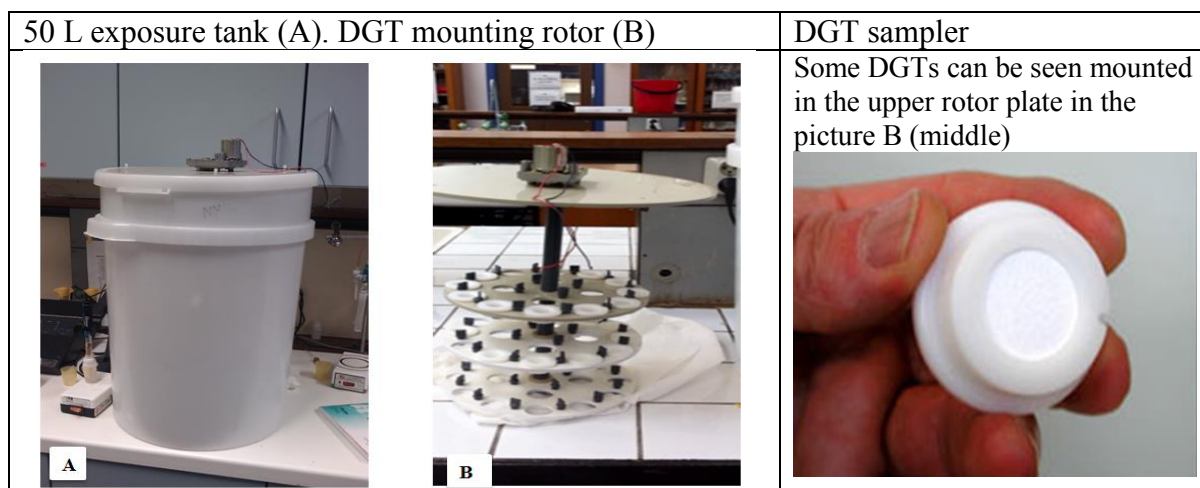
Figure 7.
The exposure tank with rotor used to test DGTs developed by Garmo, Røyset et al. (2003).

The container was filled with the test solution to a volume of 45 litres. The DGTs were mounted on the rotor disks which were immersed in the test solution. The rotor was set to a rotating speed of 6 rpm. The rim velocity at the edge of the rotor was then about 10 cm /sec to ensure a low but reproducible diffusive boundary layer (DBL) during the experiments, as the DBL is ca 0.1 at 10 cm/sec (see chapter 4.8).

2.3.2 Preparation of the DGT test solutions

Deionized water (Millipore) was used to prepare the test solutions in the exposure tank. Four exposure experiments were performed. In the first test 1 mM ammonium acetate buffer was adjusted with NaOH to pH 5.0. 1 mM NaCl was added to achieve a fixed ionic strength. This test was run over 3 weeks. Some algae growth was observed in this solution. In the next 3 experiments a 1 mM Na-acetate buffer adjusted to the desired pH was used, in order to decrease the nutrient load and thereby the risk for algae growth.

Stock solutions of AMP and IP6 were prepared and used to adjust the concentrations of AMP and IP6 in the exposure tank to approximately 25 µg P/L. This concentration was chosen to achieve a measurable concentration (ca 25 µg P/L) in the DGT extracts already after 24 hours exposure. Furthermore, the adsorbed amount after 20 days exposure would be ca 1.5 µg P, well below the adsorbent capacity of ca 7 µg. The DGTs were collected from the exposure tank at time intervals of one to three days until a total exposure period of 10 to 20 days. Concurrently the concentrations of the fractions TDP, DIP (and DOP =TDP - DIP) in the test solutions were determined.

The fractions TDP, DIP (and DOP by difference) were determined in the extract from each DGT. To assess that the uptake was linear with time, the uptake curves were plotted for collected amount of phosphorus in each fraction (DOP and DIP) versus exposure time. The D value was calculated for each measurement point according the Fick's equation (Table 7) by solving the equation for D.

The concentration of the DOP fraction in the test solution decreased slightly during the experiments. The concentration used at each time interval was therefore the average between that determined at the starting point and the one at the point when the DGT was collected. Price, Teasdale et al. (2013) experienced the same problems, and used the same calculation technique. The reasons for this problem may rely on algal growth, wall adsorption or other problems not understood for the time being.

2.3.3 Extraction of adsorbed phosphorus compounds from the DGT adsorbents

The extraction of P from the absorbent in the Fe-DGTs was performed by acids. The extractions were performed by concentrated nitric acid when using ICP-MS in earlier work (Røyset et al., 2004). This was done in the same way as for the Chelex™ DGTs. 1 mL concentrated nitric acid was added to the adsorbent and diluted to 10 mL final volume, i.e. providing a matrix of 1:10 vol/vol of nitric acid which is normally used in ICP-MS. The Fe-DGT adsorbent dissolved completely (i.e. the membrane turned from the normal brown colour to become completely clear). All adsorbed phosphate was assumed to be eluted and determined by ICP-MS (Røyset et al., 2004).

In the 2004 study above, only orthophosphate species (DIP) were examined. When determining the phosphorus fractions from the DGT tests in this study, the aim was to determine both the DIP and DOP fractions. The TDP and DIP fractions were determined using the Skalar CFA analyzer (see Chapter 3.2). To avoid possible degradation of the organic phosphorus compounds, the Fe-DGT adsorbents were not extracted by concentrated nitric acid but instead extracted using dilute sulphuric acid. This extraction was performed by placing the adsorbent membrane in 1.0 mL of 2.4 N (1.2 M) sulphuric acid overnight. This was diluted to 30 mL with deionized water in the CFA analyser auto-sampler tubes (Sarstedt 30 mL PP tubes) giving a final concentration of 0.08N sulphuric acid. Also by this procedure the ferrihydrite on the adsorbent membrane dissolved completely (i.e. the membrane turned from the normal brown colour to be completely clear). It was therefore assumed that both the inorganic and organic phosphorus compounds were eluted with 100% extraction efficiency from the adsorbent. The phosphorus fractions in the DGT extract were then determined with the CFA analyzer methods.

The same extraction method (1.0 mL 2.4 N sulphuric acid, and diluted to 30 mL) was tried for the Me-DGTs in an initial test. The TiO$_2$ Metsorb™ adsorbent did not dissolve by this acid treatment. The extraction efficiency was only 30-50% by this procedure. We therefore went back to the original extraction procedure using 1 M NaOH, as described by Panther et al. (2010). The Metsorb^{TM} adsorbent did not dissolve by this NaOH procedure either, and a check of the extraction efficiency was required to ascertain how much was extracted from the adsorbent, as also Panther et al. (2010) recommended two consecutive extractions. An extraction efficiency test by 2 repeated 1 M NaOH extraction of the Metsorb™ adsorbent was conducted (DGTs both from field and lab experiments were tested (see Chapter 4.1). The NaOH extracts were then neutralized by adding equivalent amounts of sulphuric acid. The sample was diluted to 30 mL, and more sulphuric acid was added to a final concentration of 0.08 N (30μL of 8 N sulphuric acid per 30 mL) to match with the normal 0.08 N preservation used (chapter 3.2.4). In all experiment the membranes were picked out of the solution after extraction, either by decanting the solution over to acid washed tubes or by picking up the adsorbent membrane by clean plastic forceps.

2.3.4 Calculation of the DGT-DIP and DGT-DOP concentrations from the extracts of the DGTs

The DGT fractions DOP and DIP are achieved as explained in Figure 3 and 4. First, the adsorbent membranes are dismantled from the DGTs and extracted (chapter 3.3.3). The two fractions TDP and DIP are determined independently by the CFA analyser methods (chapter 3.2), and the DOP fraction is calculated by difference as explained in Table 5. The obtained DIP and DOP fractions in the extracts are then converted to the average DGT concentration by the DGT equation in Table 6.

Table 6 contains all the input values needed for the calculation with the units used the appropriate constants, and determined values in the extracts. The development of some of the numerical values for the constant or correction functions are further discussed in chapter 4.

Table 5.

DGT fractions determined by the CFA analyser which is needed to calculate the average DGT concentration in water. The fractions in the DGT extracts are determined as TDP, DIP and DOP, which are then converted to the corresponding DGT concentrations of DGT-DOP, DGT-DIP and DGT-TDP concentrations by the parameters in Table 5.

Fractions	INPUT VALUES
In DGT extracts	**Fractions in the DGT extracts determined by the CFA analyser**
TDP	Determined with online PS digestion (as TOT-P by the SKALAR method).
DIP	Determined without digestion (i.e as PO4-P by the SKALAR method)
DOP	By difference between the fractions above (. i.e DOP =TDP - DIP)
	OUTPUT VALUES
Calculated	**Calculation of DGT concentration in water based on the DGT equation and the parameters and constant in Table 6.**
DGT-DOP	Based on the DOP fraction in the DGT extracts
DGT-DIP	Based on the DIP fraction in the DGT extracts
DGT-TDP	Sum of DGT-DIP and DGT-DOP (Note that these are DGT concentrations)

Table 6.

The input values required to calculate the average DGT water concentration of the phosphorus fractions over the deployment period based on the concentrations in the DGT extract in Table 5.

	Notation	**Default**	**Explanation**
Calculation of DGT concentration	C	ng P/mL µg P/L	$DGT\ Concentration : C = m\ /\ (t \cdot (DA/L))$
Extraction volume of DGT	V_e	30 mL	30 mL is used unless otherwise stated
Concentration in the extract	C_e ng P/mL	ng P/mL	C_e in ng P/mL determined in the DGT extract
Amount extracted from each DGT	M	ng P	m (ng) = C_e x V_e above
Deployment time	t (sec)		t (in seconds) from start to stop
Average deployment temperature.	t (^{o}C)		Average of temperature in ^{o}C at start and stop, or average temperature by a temperature logger
Diffusion coefficient (D)	D		Chosen D in cm^2/sec in the unit 10^{-6} cm^2/sec
Temperature correction factor for D	D_t/D_o		Temperature correction factor from equation 10 in Table 7 based on the average temperature
Membrane resistance factor	R	%	Correction factor from equation 10 in Table 7.
Area of the sampler window	A	3,14 cm^2	Window radius of 1.0 cm gives area: A=3.14 cm$^{2.}$
Total Diffusion length L	L	0,11 cm	L = 0,11 cm. Consists of 0.080 cm (diffusion membrane) + 0,011cm (filter) + 0.010 cm (DBL)

2.3.5 Diffusion theory

The diffusion theory for uptake of ions by DGTs is based on Fick's 1st law for steady-state diffusion in dilute solutions. The basic diffusion formulas needed to calculate the uptake by DGTs are described in the original DGT publication by Davison and Zhang (1994). These formulas were

reviewed, summarized and to some extent updated by Garmo, Røyset et al. (2003). Table 7 gives an overview of the formulas used and includes also some new formulas. The diffusive boundary layer (DBL, δ) is estimated from fluid mechanics. Garmo, Naqvi et al. (2006) made a slight modification of the DBL formula, which is used in Table 7.

The diffusion coefficients must be corrected for the DGT deployment temperature. The correction functions for temperature based on Equation 5 and 6 in Table 7 are cumbersome to work with. A simpler correction function based on only the temperature was developed in this work (see chapter 4.8 for details, and equation. 10 and 11 in Table 7). Equation 10 expresses the function for the correction factor (Dt/Do), used to adjust the D from $25^{o}C$ to the average deployment temperature. Equation 11 expresses the uncertainty of the temperature correction factor D_t/D_o per degree (^{o}C). This new function was developed in this work and can be used to estimate the magnitude of this is uncertainty component (see Chapter 4.8 for details).

The ideal situation is to determine the average deployment temperature by using temperature loggers in parallel with the DGT. This is often expensive and often not possible. The normal procedure at NIVA is to determine the temperature at start and stop of the deployment period for DGTs, and then calculate the average deployment temperature.

Buffle et al. (2007) developed an empirical equation for the relation between D and Mw, which has been is used to estimate diffusion coefficients. The use of this equation is discussed in chapter 4.3.

2.3.6 The TWA and ΔG concepts

In the DGT and passive sampling literature the TWA and ΔG concept is much used. The TWA concept refers to "Time Weighted Average Concentration". This TWA concept may be confusing, as it represents an exposure parameter in occupational health and in toxicology.

TWA is used for evaluation of a dose relationship, i.e. average concentrations multiplied by time. Also the concept "Time integrated concentration" should be avoided, as it reminds of an amount of a chemical sampled over time (integrated), which may be interpreted in similar ways as the TWA concept above.

In order to avoid the confusion the TWA concept should not be used. Instead the concept "Average concentration" over the DGT deployment period is recommended.

DGT users seem to have adopted the ΔG concept from the very early DGT publications. The main author prefer the term L instead, as this is the distance the ions need to travel over the diffusion membrane. This distance is a diffusion length (not a "membrane thickness") parameter. Therefor the term L is used throughout this text.

2.4 Field study of DGTs

2.4.1 Field Sites

The field study was performed from June to September 2011. The DGT's were deployed in three streams located at the following areas of the Morsa catchment (see Figure 5)
- The Dalen stream draining a completely forested catchment in the north of Vestre Vannsjø.
- The Støa stream draining a small watershed from an agricultural area south of Vestre Vannsjø.
- The Huggenes streamdraining a small watershed from a mixed forest and agricultural area in the south of Vestre Vansjø.

2.4.2 DGT deployments

Both the Me-DGT and Fe-DGTs were deployed in parallel in each stream. The deployment periods were 6 to 14 days. The samplers were deployed in PP plastic net tubes. After exposure the DGT's were stored prior to analysis at 4 ^{o}C for the duration of the field study, i.e. storage times were up to about 12 weeks at the longest.

Table 7.

Updated overview of the equations used to describe diffusion processes needed in the DGT work based on Garmo, Røyset, et al. (2003).

	Equations	Formulas	Denotations
1	Flux by Ficks 1st law	$Flux = \dfrac{dm}{dt} = -D \cdot A \cdot \dfrac{dC}{dZ}$	Flux of molecules (dm/dt) over a concentration gradient (dC/dz), diffusion coefficient (D).
2	Time integrated uptake	$m(t) = Co \cdot t \cdot (DA/L)$	Integration of Eq. 1. Mass uptake by time (m(t) based on D, Length (L) and cross section area (A) of the diffusion membrane
3	Average concentration	$Co = \dfrac{m}{t \cdot (DA/L)}$	Average concentration Co over the deployment period (from eq. 2)
4a	Diffusion length to be used instead of L in eq. 3	$L = (G + f + \delta) \cdot$	Components of the L term in eq. 2 and 3: the thickness of the gel membrane (G), the filter (f) and the diffusive boundary layer (δ)
4 b	D coefficient derived from eq. 3	$D = \dfrac{(m \cdot (G + f + \delta))}{t \cdot Co \cdot A}$	Calculation of D from laboratory experiments based the amount o analyte (m at time t) at each measurement point at the water concentration Co
4c	DGT uptake rate in volume per time unit based on the DA/L term	$DGT\ uptake\ rate = \dfrac{D\,A}{L}$ Units mL/sec, mL/h, mL/day (24 h)	The uptake rate is the volume of water "diffusively emptied" of ions per time unit. At D = $5 \cdot 10^{-6}$ cm^2/sec, L=0.011 cm, A=3.14 cm^2, the uptake rates are ~0.5 mL/h or ~12 mL/24h at 25 ^{o}C.
5	Temperature correction of D (Stokes-Einstein)	$\dfrac{D_t \eta_t}{T_t} = \dfrac{D_O \eta_O}{T_O}$	Absolute temperature (T), viscosity (η) and D by the Stokes-Einstein equation
6	Temperature correction of viscosity	$\log \dfrac{\eta_o}{\eta_t} = \dfrac{1{,}37023(t-25) + 0{,}000836(t-25)^2}{109 + t}$	Adjustment for the change in viscosity of water with temperature. Temperature correction of D require combination of eq. 5 and 6 (as in eq. 10.)
7	Diffusive boundary layer (DBL) thickness	$\delta = 3.3 \cdot \sqrt[3]{\dfrac{D}{v}} \cdot \sqrt[2]{\dfrac{vx}{U}}$	v kinematic viscosity, x distance from the leading edge, U flow velocity of the free solution, D is the diffusion coefficient.
8	Reciprocal mass plot for DBL estimation	$\dfrac{1}{m} = \dfrac{(G + f)}{D \cdot Co \cdot A \cdot t} + \dfrac{\delta}{D \cdot Co \cdot A \cdot t}$	Calculation of DBL (δ) from measurements with DGTs with at least two gel thicknesses (reciprocal plot method)
9	Buffle's equation 25°C	$D = \dfrac{2.84 \times 10^{-5}}{\sqrt[3]{M_w}}$ (cm^2/sec)	Estimate of D from molecular weight (Mw) based on the work by Buffle et al. (2007)
10	Temperature correction of D	$\dfrac{Dt}{Do} = 0{,}0001947 \cdot t^2 + 0{,}01716 \cdot t + 0{,}4492$	Based on Eq. 5 and 6 above. t is temperature in ^{o}C. (based on ch, 4.7 in this work)
11	Uncertainty of Dt/Do	$Uncertainty\ (\%) =$ $0.000419 \cdot t^2 + 0.054475 \cdot t + 3.765$	Uncertainty (%) in the Dt/Do factor (Eq.. 10) for correction of D. t is the temperature
12	Membrane resistance R	$R = D_{DGT} / D_w$	Membrane resistance R
13	pH correction of D for orthophosphate	$D = -0.15 \cdot pH + 6.62$	Correction of D for pH for the orthophosphate species in the APA membrane (this work) at R=30%

3. RESULTS AND DISCUSSION

This chapter starts with an assessment of the analytical performance parameters which are available from the experiments with the DGT samplers:

- The extraction efficiency of the P compounds collected on the DGT adsorbent membranes
- The sampling precision in in the field, of both the Fe- and Me-DGT samplers for the collection of the TDP, DIP and DOP fractions
- Comparison of uptake of the TDP, DIP and DOP fractions by the Fe- and Me-DGT samplers in the field.
- Estimates of the diffusion coefficients (D) of the model compounds AMP and IP6 based on results from laboratory experiments.
- Estimates of the diffusion coefficients for the DIP and DOP fractions found in natural waters based on literature values, as well as by comparisons between data from stream water samples and DGT data from the field study.

The following topics are also discussed and assessed

- Evaluation of source patterns in water from forest and agricultural soils
- Review of the temperature correction function for D
- Examination of the uncertainties of DGT measurements
- Summary of important performance properties of the DGT samplers

3.1 Estimation of extraction efficiency

3.1.1 Fe-DGTs

The Fe-DGTs adsorbents consisting of ferrihydrite ($Fe(OH)_3$) are extracted by dilute sulphuric acid (2.4 N). The iron hydroxide in the adsorbent dissolves completely. The extraction efficiency with 2.4 N sulphuric acid is therefore considered to be 100%.

3.1.2 Me-DGTs

The extraction of P from the Me-DGT using the same sulphuric acid solution (2.4 N) as for the Fe-DGTs gave recoveries of only 30 to 50%. The original extraction of Me-DGT using 1 M NaOH in 2 consecutive steps was therefore used (Panther et al. (2010), see also Chapter 3.3). The results in Figure 8 and the summary in Table 8, shows an extraction efficiency of the first extraction of about 87 % (13% in the second step) for both the Me-DGT-TDP and DGT-DIP fractions. The same extraction efficiency of 87% was achieved in the laboratory study with IP6.

The extraction efficiency is very linear over the whole concentration range from 1 to ca 300 µg P / L in the DGT extracts (with high correlation coefficients of R^2 of 0.97 to 0.98). The slopes are also very similar, i.e. 0.138 and 0.133. The extraction efficiency of the first extraction step is thus from 0.878 (1:1,138) to 0.882 (1:1,133), i.e. about ca 88 % of the first step. The precision achieved for the first extraction in the laboratory test of IP6 were very good (1%), while somewhat poorer (4%) for the field samples.

The LMW molecule IP6 showed extraction efficiency of 88% with high precision. The good extraction efficiency of the DGTs from the field is also promising. These compounds may cover a molecular weight range from 300 up to at least 5000 and maybe up to ca 10 000 Da. The good extraction efficiency of these compounds is promising as the DGT Metsorb™ sampler then can be used for field sampling with high and reproducible extraction efficiency in the laboratory.

If the absorbent is only extracted once a correction factor of 1.13 (i.e. 100/88) can therefore be used. If more accurate results are required two extractions are recommended.

Another way to increase the extraction efficiency from Me-DGTs is to increase the strength of the eluents (to 5-10 M NaOH, or use other eluents such as concentrated NH_3 (ca 15 M), or a strong not oxidizing acid such as HCl)). This approach was not studied here, but can be a topic for future research.

Table 8.
Extraction efficiencies obtained for the Metsorb™ based DGTs (Me-DGT).

Extraction efficiency test Me-DGT	N	Extraction efficiency %	Standard dev.	RSD, %
Field test (Støa, Huggenes and Dalen)	25		4	4 %
Laboratory test of IP6	10	88	1	1 %

Extraction efficiency DGT-TDP (field)	Extraction efficiency DGT-DIP (field)
Me-DGT DGT-TDP Extraction efficiency - field samples $y = 0{,}133x$ $R^2 = 0{,}9703$	Me-DGT DGT-DIP Extraction efficiency $y = 0{,}138x$ $R^2 = 0{,}982$
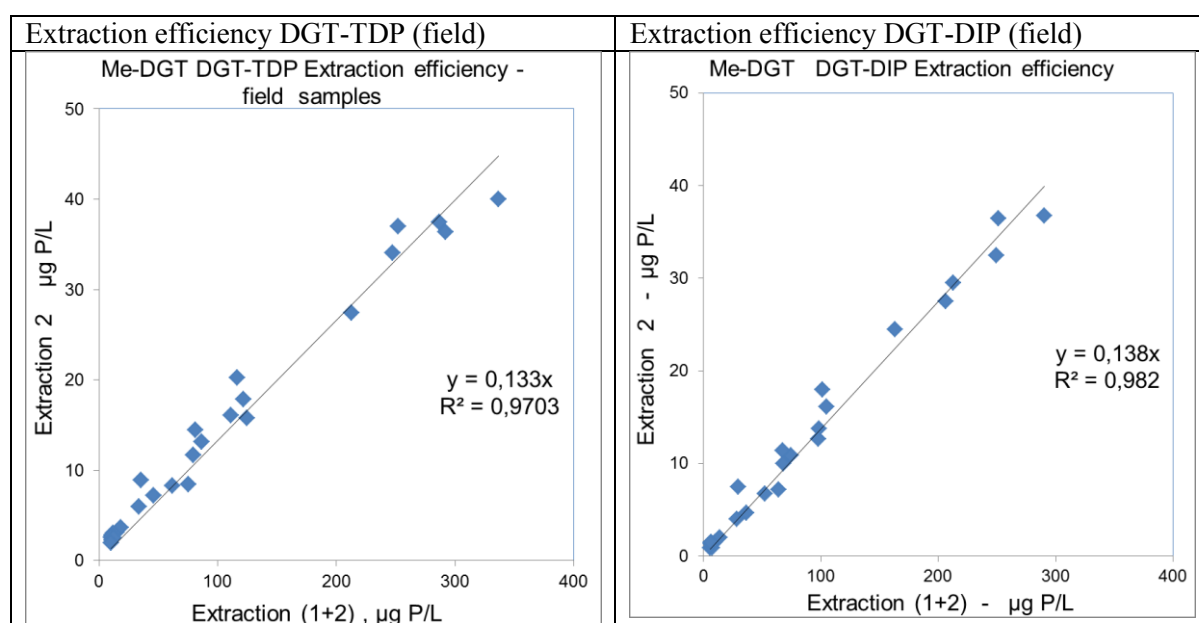
Figure 8.
Extraction efficiencies obtained by two consecutive extractions of the Me-DGT by 1 M NaOH. The data are from the field study described by Parekh (2012). The extraction efficiencies are calculated from the concentrations in the DGT extract (the calculation to DGT average concentrations will not influence the extraction efficiencies).

3.2 Estimation of Diffusion coefficients from the laboratory experiments

3.2.1 Experimental data

Three laboratory experiments were performed with AMP and one with IP6. The uptake curves from the 4 laboratory tests are shown in Figure 9 graph A to D. The D-values are shown Table 9 and 10.

The best results were achieved with the Fe-DGT sampling of AMP over 20 days (graph A in Figure 9). Notably, there are no measurable amounts of free orthophosphate in the extract (DIP fraction) in the extract from the sampler (points labelled green in A). The extraction of the Fe-DGT adsorbent was performed with 2.4 N sulphuric acid and diluted to 0.08 N sulphuric acid prior to analysis of

TDP and DIP fractions of the DGT extract. This indicates that the AMP remains in organic form on the adsorbent, is eluted as AMP, and remains as AMP in the 0.08 N sulphuric acid until analysis. Similar results using the Fe-DGT were achieved with IP6 (graph D). Also in this experiment no measurable amounts of DIP were detected. This indicates that all adsorbed IP6 is extracted as IP6. I.e. No degradation or release of any of the 6 phosphate groups on the IP6 molecule seem to occur

The high stability of AMP and IP6 on the adsorbent during the extraction step and during storage is very important. This indicates that the adsorbed DOP compounds do not degrade during sampling, extraction (2.4 N) and storage in the 0.08 N sulphuric acid. The good and reproducible extraction efficiencies of the Me-DGTs from the field study (chapter 4.1) indicate the same. AMP is considered to be very stable in acid solution (1 year, NIVAs Tot-P procedure D3), which also is stated by Moorleghem et al (2013) for IP6.

Figure 9.

Uptake curves of AMP and IP6 on Fe- and Me-DGTs performed in laboratory studies by Mohr (2010) and Parekh (2012). The concept TDP corresponds to Total P and DIP to PO_4-P.

Two experiments (graph B and C) showed a larger deviation from the linear TDP adsorption curve, especially for the sample collected after 3,5 and 7 days Also some degradations of AMP appear to have occurred, as low but measurable amounts of the DIP fraction (labelled green in Figure B and C). These d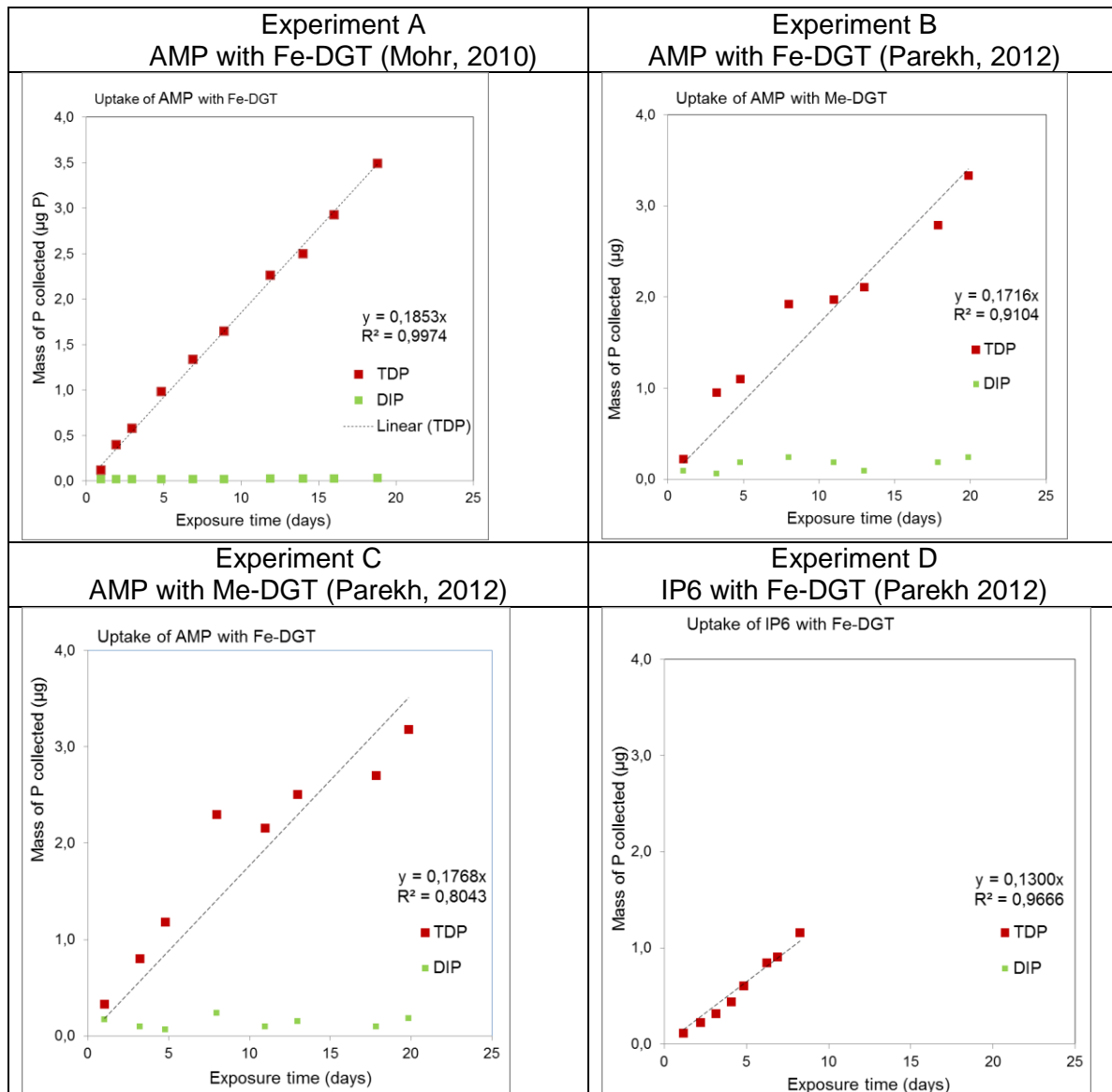eviations may have been caused by an extended storage time prior to detection, allowing a slow degradation of the AMP. However, the linear regression curves is very similar for all 3 AMP experiments, with coefficients between 0.17 and 0.18 (see graphs A, B and C). This indicates that the point between 10 and 20 days (which controls the slopes,), are correct, and that the adsorption is quantitative for AMP on both the Fe-DGT and Me-DGT samplers), and that the capacities of the sampler adsorbents were not overloaded. The maximum adsorbed P was ca. 3.5 µg P, while the capacities are ca 7 µg (Fe-DGT) and 37 µg P (Me-DGT), (see Table 25, chapter 5). The linear uptake curve for IP6 (graph D) indicates the same.

The diffusion coefficients derived from data in Figure 9 are shown in Table 9 and 10. The RSDs of the experiments range from ca. 3 % to 22 %. The average RSD for the 3 AMP tests were 14 % (3.16 (0.44), Table 10). This uncertainty is acceptable as these values were achieved in the first laboratory tests. It is likely to achieve lower uncertainty as we gain more experience on the test conditions (such as the very good precision of the test A for the Fe-DGT).

Table 9.
Diffusion coefficients calculated from the data in Figure 9. The diffusion coefficients are adjusted to 25°C by the Stokes Einstein equation (eq. 6 in Table 7) according to the average temperature (^{o}C) during the exposure period.

Experiment		A	B and C	B	C		D
Compound		AMP		AMP	AMP		IP6
Sampler		Fe-DGT		Me-DGT	Fe-DGT		Fe-DGT
	Time,	D	Time	D	D	Time	
	days	10^{-6} cm^2/sec	days	10^{-6} cm^2/sec	10^{-6} cm^2/sec	days	$\cdot 10^{-6}$ cm^2/sec
	0.9	2.10	1.0	3.36	4.26	1.1	1.08
	1.9	3.38	3.2	4.08	3.29	2.2	1.11
	3.0	3,35	4,8	3,02	3,14	3,1	1.11
	4.9	3.44	8.0	3.48	4.07	4.1	1.19
	6.9	3.30	11.0	2.65	2.84	4.8	1.38
	8.9	3.22	13.0	2.48	2.90	6.2	1.54
	11.9	3.31	17.9	2.50	2.39	6.9	1.50
	14.0	3.19	19.9	2.68	2.54	8.2	1.60
	16.0	3.32					
	18.8	3.37					
Average D		**3.32**		**3.03**	**3.18**		**1.31**
Std dev.		**0.081**		**0.57**	**0.67**		**0.214**
RSD		**2.4**		**18.7**	**21.7**		**16.3%**
pH		5.0 (0.2)		5.5 (0.2)	5.5 (0.2)		5.5 (0.2)
Temperature ^{o}C		22		22-23	22-23		22-23
MSc work		Mohr (2010)		Parekh (2012)			

Table 10

Diffusion coefficients (D) obtained or AMP and IP6 from the laboratory experiments shown in Figure 9 and from estimates based on Buffle's equation.

Organic P	DGT (experiment)	Mw	pH	Diffusion coefficient (D) (std. dev.) 10^{-6} cm^2/sec	D estimated by Buffle $\cdot 10^{-6}$ cm^2/sec
AMP	Fe-DGT (A)	347	5.0	3.32 (0.08)	4.0
AMP	Fe-DGT (B)	347	5.5	3.18 (0.67)	4.0
AMP	Me-DGT (C)	347	5.5	3.03 (0.57)	4.0
IP6	Fe-DGT (D)	736	5.5	1.31 (0.21)	3.2
AMP	Average (A, B, C)	347	5.0 and 5.5	3.16 (0.44)	4.0

Table 11.

Estimates of diffusion coefficients (D) from 10 to 1 000 000 Da, based on data by Logan (2012), Buffle et al. (2007) and the compilation by Li and Gregory (1974). Some experimental based DGT diffusion coefficients are included, obtained in this for AMP and IP6, and the orthophosphate species as well as some other small ions (to examine how good the prediction of Buffle's equation is for ions in the low Mw range).

Analyte	Molecular weight	Molecular diameter Logan (2012)	Diffusion coefficients			
			Logan (2012)	Buffle (2007)	Li and Gregory (1974)	DGTs
	Mw	Md	10^{-6}	$\cdot 10^{-6}$	$\cdot 10^{-6}$	$\cdot 10^{-6}$
	Da	Nm	cm^2/sec	cm^2/sec	cm^2/sec	cm^2/sec
	10	0.23	22			
F	19			11	14.6	
Na	23			10	13.3	
Small ion (Co^{2+})	59			7.4	6.99	6.14*
	100	0.62	7			
Medium ion (Cd^{2+})	111			6.0	7.17	6.10*
Large ion (Pb^{2+})	207			4.9	9.45	10.0*
Organic acid	300			4.5		
Organic acid	400			4.0		
Organic acid	500			3.7		
Fulvic acid	1 000	1.3	2.5	2.9		
Humic acid	2 000			2.5		
Humic cmpds	5 000			1.7		
Humic cmpds	10 000	2.85	1.1	1.4		
Large molecules	20 000			1.1		
Large molecules	50 000			0.80		
Large molecules	100 000	6.2	0.5	0.64		
Large molecules	1 000 000	13.2	0.25	0.30		
AMP	347			4.0		3.2 ***
IP6	736			3.2		1.3 ***
H$_3$PO$_4$	98			6.3		
H$_2$PO$_4^-$	97			6.3	8.46	6 ±0.5**
HPO$_4^{2-}$	96			6.3	7.34	
PO$_4^{3-}$	95			6.3	6.12	

*Garmo, Røyset et al.,(2003), **) Røyset, Sogn et al (2004), ***This work Parekh (2012) and Mohr (2010).

3.3 Estimation of diffusion coefficients

3.3.1 Data sources for diffusion coefficients (D)

The diffusion speed of ions and molecules is water depends on several factors such as molecular weight, charge, size and shape (spherical vs linear) etc. Diffusion theory is an old science developed by the works of Fick, Stokes, Einstein etc, from the 18th and early 19th century, and is described in monographs of chemical and physical engineering and transport processes such as by Cussler (2009) and Logan (2012).

Li and Gregory (1974) have compiled diffusion coefficients for inorganic ions in water. These data for the free diffusion of ions in water are based on limiting conductance data as well as other data sources. This dataset is probably one of the best compilations available for inorganic ions, and has been used by DGT researchers. Some important numbers are given in Table 11. Li and Gregory's data for the orthophosphate species are discussed in chapter 4.3.5.

DOM compounds have molecular weights from LMW <1kDa to high molecular compounds up to 1000 kDa. Except for the review by Buffle et al (2007) discussed below, few good compilations are available for D values of molecules of relevance for DOM or DOP compounds in water. Logan (2011, and 2012) presents some equations for the estimation of D as shown in Table 12.All are based on empirical relations between D and Mw for different types of macromolecules in the Mw ranges from 1000 to 1000 000 Da.

Table 12.
Overview of equation to estimate diffusion coefficients based on molecular weight for macromolecules based on (Logan 2012). All the equations were given at 20°C. The Beckett equations have not specified temperature, but are probably at room temperature (i.e. closer to 20 than to 25°C).

Application area	Diffusion coefficient D, cm^2/sec	Abbre-viation	Tempe-rature	Source
Spherical molecules with known radius	$D = \dfrac{k_B \times T_T}{6\,\pi\,\mu\,r}$	Stokes Einstein		Logan (2011)
Molecules in the range 200 to 100000 Da	$D = 2.84 \times 10^{-5} \times M_w{}^{-1/3}$	Buffle2007	20°C	Correlation by Buffle et al. (2007)
Molecules in the range 200 to 100000 Da	$D = 3.3 \times 10^{-5} \times M_w{}^{-1/3}$	Buffle1988	20°C	Correlation by Buffle et al. (1988)
Proteins Mw>1kDa	$D = 2.74 \times 10^{-5} \times M_w{}^{-1/3}$	Frigon1983	20°C	Frigon correlation Logan (2012)
Dextrans Mw >1kDa)	$D = 7.04 \times 10^{-5} \times M_w{}^{-0.47}$	Polson1950	20°C	Polson correlation, Logan (2012) *
Humic and fulvic acids	$D = 1.42 \times 10^{-4} \times M_w{}^{-0.42}$	Beckett1987	Not given	Beckett correlation Logan (2012) *
Humic and fulvic acids	$D = 6.00 \times 10^{-5} \times M_w{}^{-0.42}$	Beckett1987 modified	Not given	Beckett correlation Beckett et al 1987 *

*) Original publications: Beckett et al.(1987), Frigon et al. (1983), Polsen, A. (1950).

Some confusion has appeared with respect to the temperatures at which the equation should be used. Buffle has referred to 20 and 25°C for his 1988 and 2007 equation, but it appears that both are given at 20°C (Buffle 2014). The Beckett equation lack temperature specification, but is probably close to 20°C. Thus, the equation in Table 12 is expected to be at 20°C.

Logan (2012) recommends the Stokes Einstein equation given in Table 12, as a general approach for estimating D in water. The Stokes Einstein equation is a basic physical chemical relationship

developed for spherical molecules and requires the Boltzman constant (k_B), the absolute temperature (T), dynamic viscosity (μ), and molecular radius (r). The equation works best for larger molecules above 1 to 10 kDa (Logan 2012). Accurate data for k_B, T and μ are available. However, the molecular radius (r) is not readily available for macromolecules like DOM or DOP compounds in water. In water, the radius has to be converted to the hydrodynamic radius including the thickness of the water cap around the ionic molecule. This parameter requires careful evaluation for dissolved ions in water especially for charged (ionic) molecules below 10000 Da. Although the molecular weight range of the DOM/DOP group both can be estimated and even determined, the hydrodynamic radius including the water cap of the individual DOM or DOP compounds may change with ionic strength, pH, ionic shape and other parameters. Thus the uncertainty of the input parameter for the hydrodynamic radius may introduce large uncertainties in the estimate of D for DOM/DOP compounds when using Stokes Einstein. The Stokes Einstein equation has thus several limitations when used for estimating D for DOM or DOP compounds especially at Mw below 10000 Da.

DGT researchers have most often used the work by Buffle and co-workers for estimating D values for humic compounds. Buffle developed a semi empirical relation between Mw and D for some humic molecules and other compounds from 200 to ca 100 000 Da. Buffle developed his equation in two versions, the one in his monograph from 1988 (Buffle et al. (1988) and in a publication from2007 (Buffle et al. (2007), which for simplicity is referred to as the Buffle1988 and 2007 equation in Table 12. These 2 equations deviate only in the constant of 3.3, and 2.84. The reason for this is unfortunately not mentioned in the Buffle et al. (2007) publication. However, the original equation (Buffle et al. (1988) also included the so-called frictional ratio factor:

$$D = \quad 3.3 \cdot 10^{-5} / \left(\frac{\phi}{\phi o}\right) \quad \cdot M_w^{1/3} \qquad \text{Original equation in cm}^2/\text{sec by Buffle et al. (1988).}$$

The difference is due the use of the frictional ratio, which is expressed as the ($\phi/\phi o$) term (Buffle (2014), pers. comm.). According to Buffle this frictional ratio is always larger or equal to unity, (i.e. $\phi/\phi o > 1.0$), but increases when molecules are non spherical and/or hydrated, which is the case of fulvic and humic compounds. According to Buffle the frictional factor goes asymptotically to 1.00 as Mw goes to zero (0). Few numbers are available, but Buffle mentions a number of 1.12 at Mw of ca 2000. The factor increases to ca 2.0 at 1000000Da. Thus when the value of ($\phi/\phi o$) is included in the denominator of the equation, the net effect is that the constant of the equation decrease with Mw. This is the case in in the Buffle2007 equation where the factor is 2.84. This is due to that he included a frictional ratio of 1.16 (3.3/2.84) corresponding to a Mw between ca 2000 to 5000 Da. The Buffle1988 equation has been used by DGT researchers until now (Zhang et al (1999), Jones, Chen, Zhang et al 2014)). This has introduced confusion on which is the most appropriate for humic compounds. Based on the new information above the Buffle2007 equation is recommended for humic compounds when the frictional ratio is not known, but using a default factor of ca 1.16 (Buffle (2014), pers. comm). Further conclusions cannot be drawn at present.

Figure 10 show the curves obtained by the D vs molecular weight (Mw) based on the equation in Table 12. The Beckett 1987 equation for humic compounds cited by Logan (2012) seem to be an obvious outlier, as the D values are about a factor 2 higher than by the other equations. When checking this against the original publication (Becket et al 1987) it appears that this affected by an interpretation error by Logan. This equation was developed for humic compounds based on FFF (Field Flow Fractionation) measurements with Mw derived from the highest peak of the FFF fractograms. Becketts formula was based on ca 8 humic and fulvic acids in the range 800 to ca 4000 Da, but included also calibration with polystyrene (PS) standards in the range 5000 to 100000 Da. However, after having re-calibrated his fractograms, Beckett modified his formula. This (Beckett 1987 modified) seems to agree with the other data sources (this modification has not been noticed in the review by Logan (2012)) (See Figure 10).

Figure 10.
Curves of D vs Mw obtained by the equations shown in in Table 12 in linear - log scale (upper) and in log - log scale (lower).
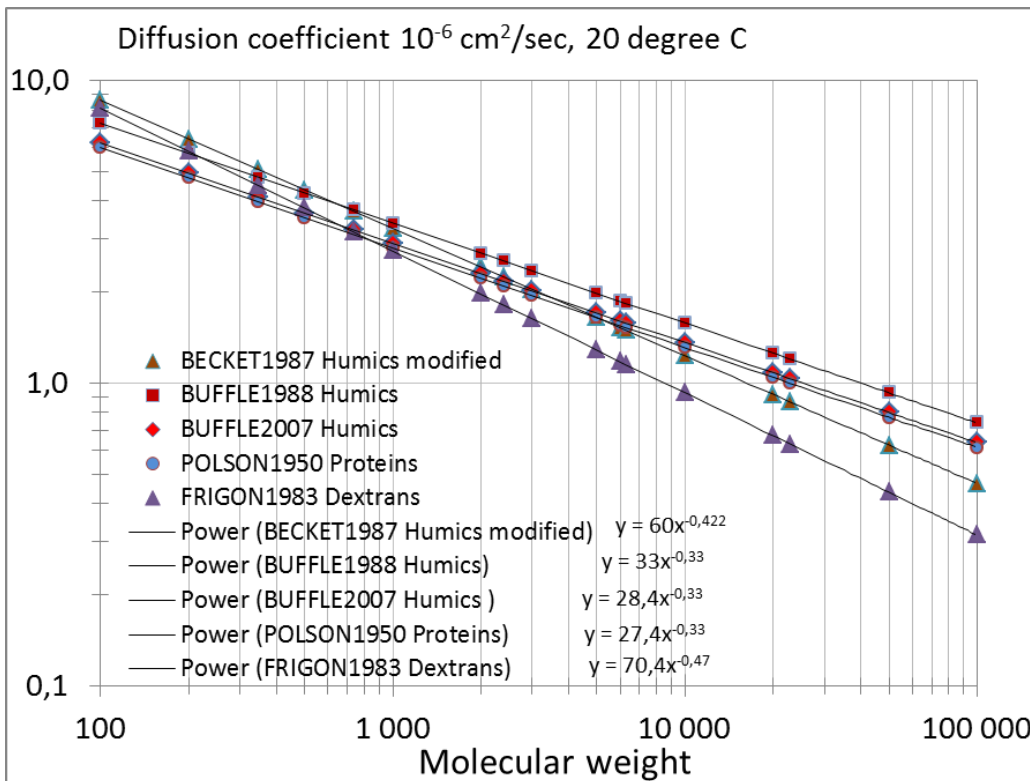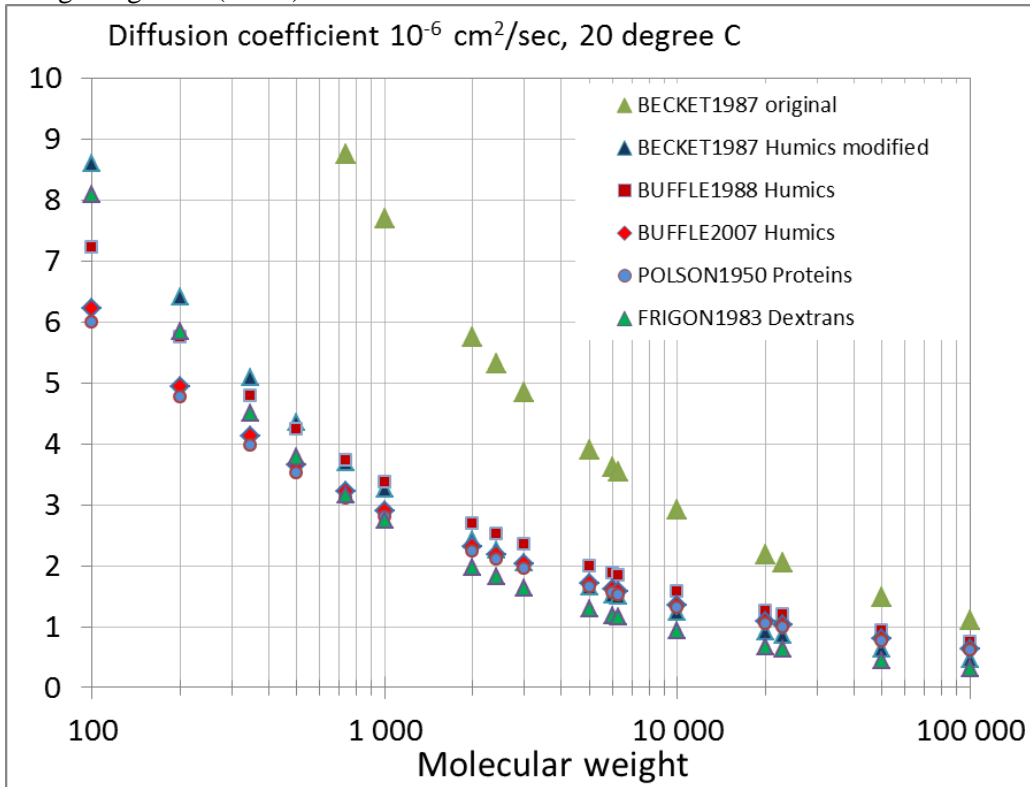
The curves in Figure 10 show that the Frigon1983 curve for proteins has the largest deviation. At Mw above 10000 the D values are lower than the other curves, probably caused by that proteins are more charged than the dextrans and humic compounds, which reduce the mobility, and thereby the diffusion coefficients. Also the Beckett1987 modified curve has a pattern similar to the Frigon1983 curve, but with larger deviation at Mw above 10000 Da. However this latter curve is based mainly on a few humic compounds (and some polystyrene standards), so larger uncertainty in this curve may be expected. The Polson1950 and Buffle2007 curves are very similar with deviations of only ca 4 % (factors 2.74 compared to 2.84). The Buffle1988 and Buffle2007 equations are parallel but has the 16 % deviation due to the frictional factor discussed above.

The Buffle2007 equation is located in the middle of these of these equations. As the frictional factor goes towards 1.0 at lower Mw, the equation with a factor between 2.84 and 3.3 may be more appropriate in the low Mw range (<1000). At present the Buffle2007 equations seem to be best suited for humic DOM and DOP like compounds from 1000 -10000 Da. This equation is therefore used in the further discussions.

Table 13 shows the deviation between the highest and lowest estimates obtained by the equation of Table 12, is in the order of 0.4 to 0.8 D units (10^{-6} cm^2/sec) shown in Figure 10. Based on these equations the uncertainty (based on Highest – Lowest divided by the average D) of the prediction of D is lowest at a Mw of 500 /ca 20%) and increases to ca 50% around 10000 Da.

Table 13.
Overview of the Diffusion coefficient (10^{-6} cm^2/sec) obtained from the different equations displayed in Figure 10. The lowest, highest, average and difference (Highest-Lowest) values for D are listed. This is based on the raw data shown in Table 27 in Appendix A.

Molecular weight, Mw	Lowest D	Highest D	Average D	Difference between Highest – Lowest compared to the average D	
				In D units	In %
100000 Da	0.31	0.74	0.57	0. 44	77
10000 Da	0.9	1.6	1.3	0.7	54
5000 Da	1.29	1.99	1.7	0.7	41
3000 Da	1.6	2.35	2.1	0,7	33
1000 Da	2.7	3.38	3.1	0.7	23
500 Da	3.52	4.36	4.0	0.8	20

Molecular diameter, molecular weight and D
Figure 11 presents relations between D vs molecular weight and molecular diameter vs , molecular weight . The D and the molecular diameter is inversely proportional.

The most important pattern to notice in Figure 11 with respect to DGT's, is the relation between the molecular diameter and the molecular weight. Even at a molecular weight of 100 000 Da the molecular diameter for a spherical molecule is not larger than about 6 nm. The conventional DGT membranes have pore size of 5 -10 nm. Thus, molecules up to a molecular weight of 100000 Da are small enough to pass a conventional 5-10 nm pore-size DGT APA membrane, although with reduced diffusion rate.

The molecular diameter can be estimated based on the atomic volumes of the individual atoms in the molecule as recommended by Logan (2012). This requires then the atomic ratios and an estimate of the formulas for the molecules which usually is not available for DOM/DOP compounds. If the molecular radius is estimated, the D can be calculated by the so-called Wilcke Chang correlation (Logan (2012)). However this topic is too comprehensive to be discussed here.

Figure 11.
Relation between Molecular diameter and D(in a log - log scale) and Molecular diameter and Molecular weight (linear log scale). The D values are based on the Buffle 2007 equation, while the molecular diameter vs Molecular weight data is from Logan (2012).
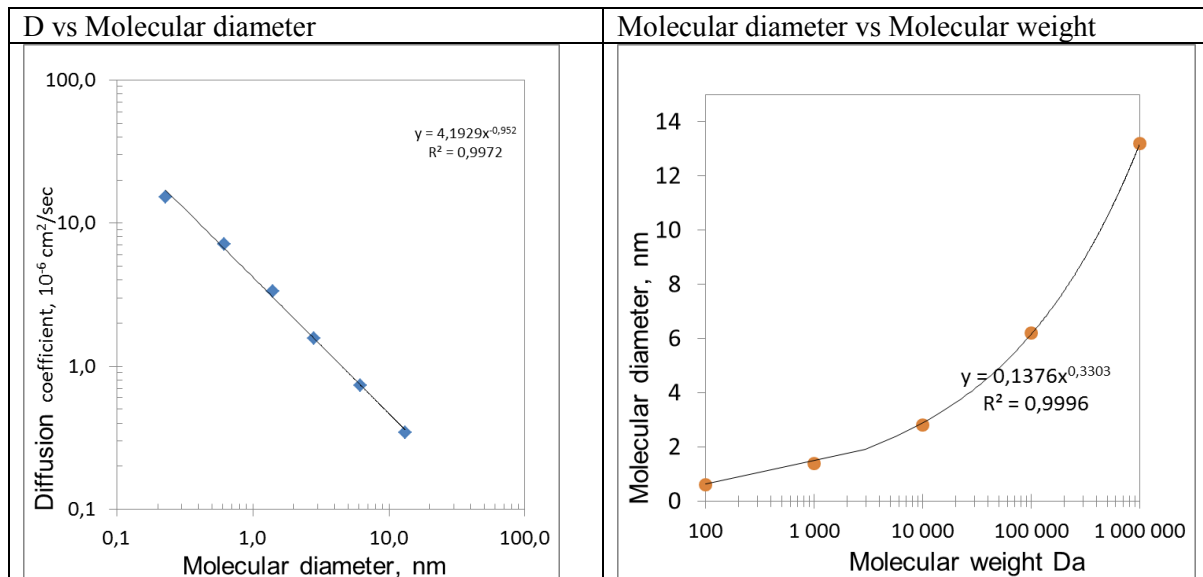
The conclusion with respect to diffusion coefficients (D) so far are:
- D values for the orthophosphate species are based on Li and Gregori's compilation from 1974. A correction for the effect of pH is introduced in this work.
- At present useful estimates for D values for DOM/DOP compounds can be achieved by the Buffle's equation in the Buffle2007 version.
- The D values for DOM/DOP compounds obtained must be corrected for factors influencing the D in water and in the membrane, as discussed in the following chapter 4.4.

3.3.2 Understanding membrane resistance

The diffusion of ions and molecules in an aqueous solution depends on several parameters such as the molecular weight, molecular size, shape and charge. Generally the diffusion rates in polar solute such as water decreases with the ionic charge, as the interaction with the water molecules increases with charge. Buffle's equation and the other equations shown in Table 12, was developed to estimate the diffusion coefficient of free ions or molecules in water. The interaction with the resistance to movement induced by a membrane is not included in these models.

The molecular diffusion though the pores of the DGT membrane depends on how molecules interact with the properties of the membrane (poresize, form of pores, charged sites, etc). Both the membrane properties and the diffusion of molecules and ions in water depend on properties of the solution (pH, ionic strength, etc). These affects the diffusion rate in the membrane compared to those tabulated for free diffusion in water. At present no models predict how all these properties and factors influence the D in the DGT membranes. The simplest approach to handle these effects, is to pool the influence of these parameters into one factor called "**the membrane resistance R**". For practical purposes the tabulated or calculated diffusion coefficients above must be adjusted with the **membrane resistance R.** The mathematic relations needed for the R factor is described at the end of this chapter. First we need to discuss some of the basics behind the membrane resistance factor used in passive sampling.

The membrane resistance is a general term which is used for all types of passive sampler, and was introduced to the passive sampler community during the 1970thies in order to calculate uptake rates for gasses in air by the use of macroporous diffusion membranes. Passive samplers for gasses in air and for compounds in water have basically two types of membranes or diffusion barriers:

Passive sampler for gasses in air:
- Open tubular samplers of the Palmes type or shorter tubes with a diffusive filter barrier at the end of the tube. The diffusion occurs in the open tubular area behind the protection (wind) filter. Except for the diffusive barrier filter, these have no membranes or pores to consider.
- Samplers with macroporous membranes (badges or similars) where diffusion occur in the pores of an often fairly thin membrane with large surface area. Several such samplers are available with different designs from flat membranes to radial membranes.

Passive sampler for compounds in water
- The DGT type where diffusion occurs in the water filled pores of a gel type membrane.. These membranes can be interpreted and understood by the properties of macroporous membranes.
- Diffusion and permeation through a thin organic polymeric ("plastic") membrane to an underlying adsorbent such the SPMD or POCIS type. Some membranes may possess small water filled pores, which makes the process a mixture of diffusion in water and permeation in the plastic membrane. Moreover, the diffusive boundary layer (DBL) on top of the adsorbent add to the total diffusion path (length) and has to be taken into account.
- Only a solid adsorbent of a polymeric material exposed to water. The diffusion is through DBL layer of water (DBL) at the adsorbent surface.
- For the two latter types the DBL layer is a considerable part of the total diffusion length (eq. 3 and 4 in Table 7). As the DBL layer depends on the water velocity, the uptake rate is more affected by the water velocity rate than for the DGT type samplers with a relatively long diffusion path compared to the DBL.

In the macroporous passive sampler membranes the molecules (gases or ions) have to travel through the pores. Two parameters are important to consider: the porevolume and the turtuosity. The turtuosity is a parameters which describe how "bended or twisted" the pores are compared to straight tubular holes in the membrane. The turtuosity is expressed as an unitless number from 1

and upwards, and can be used to calculate the effective distance the molecules have to travel through the pores before being trapped by the adsorbent.

This can be converted to an "effective diffusive length" of the macroporous membrane (an example is a sampler where a ca 2 mm macroporous membrane corresponds to a diffusion length of 18 mm). These factors are then combined to a sampling rate which is specific for the geometry and membrane material of the sampler. The down side of this approach is that all uptake rates have to be determined experimentally for all the individual gasses. This is almost the same approach needed for the passive samplers used for organic compounds in water such as the SPMD, POCIS and similar systems. The uptake rate for all compounds has to be determined experimentally. We cannot rely on the diffusion coefficients in water for these ions as the diffusion mechanism is a mixture of diffusion in the DBL water layer at the membrane/adsorbent surface and adsorption at the surface, permeation into the adsorbent or permeation through the membranes (POCIS) into the adsorbent. This ends up in time consuming calibration work for uptake rates of these types of passive samplers.

The benefit of the DGT sampler is the very simple design which permits the calculations of the uptake to be based almost exclusively on diffusive theory based on Fick's 1st law. Thus, a common understanding has been that only the diffusion coefficient of the ion in water is needed. The remaining corrections can be done by basic diffusion theory based on the concepts and equations needed shown in Table 7. This is based on the understanding that the DGT membrane is a "stationary region" of water. However, also the DGT membrane has a pore volume and turtuosity. The simplest way to estimate the porevolume is to determine the water content of the membrane. This has been done by Zhang and Davison (1999). Table 14 shows pore volumes of common DGT membranes based on this approach with reported porevolume values of 98% (AGE), 95% (AGE) and 84% (CGA). The volume of water for "free diffusion" is thus reduced. The pores are far from being straight, and the "effective diffusion path" may increase (i.e. the L - term). This turtuosity effect of DGT membrane have not been determined earlier. As long as the diameter of the pores in the membrane are much larger than the diameter of individual metal ions of around 0.1 nm, these membrane resistance effects are hardly detected in the normal AGE and APA DGT membranes. This is probably the reason why this topic has not been much discussed by DGT researchers when they have only worked with the collection of metal cations.

Many DGT researchers have chosen a workaround by using concepts like "The DGT effective diffusion coefficient" or "DGT based diffusion coefficients", which actually conceals the membrane resistance problem. This also makes it difficult to separate out the effects caused by adsorption efficiency of the compounds at the adsorbent, and from those caused by membrane resistance effects. In earlier works Garmo and Røyset et al (2003) found that the D for a number of ions were 5 -15 % lower in the APA membrane compared to the tabulated values by Li and Gregory (1994). They explained this at that time as a mixture of effects: reduced desorption efficiency, reduced adsorption efficiency at the adsorbent, back diffusion, as well as "membrane " effects. Similarly, Zhang and Davison (1999) found that the D for Cd and Cu was reduced by ca 30% in the constrained CGA membrane. This reduction for D of 20 to 40 % was also found for a number of metal cations in a study of the CGA membrane (Røyset and Garmo, 2002). Also the tabulated values of DGT based diffusion coefficients suffers from these weaknesses, i.e. they are only relevant for the APA membrane.

Thus, also DGT researchers will need to cope with membrane resistances. In the studies of the collection of molecules with Mw from 300 to at least 10000 Da for the DOM and DOP fractions in water, the R factors need to be evaluated and estimated. This topic also becomes particularly important when the DGT research now is moving towards the collection of organic compounds such as antibiotics, pharmaceuticals, personal care products (PPCP) etc. (Jones, Chen et al (2014)).

They used a correction for porevolume of the membrane by an equation based on Achies law

$$D_{DGT} = D_W \times \varepsilon^m$$

The factor ε is the porosity of the membrane and *m* is Archies law coefficient (ranging from 1.5 to 2.5 for porous media (see http://en.wikipedia.org/wiki/Archie's_law). They used the AGE membrane and proposed a value of 0.98 for ε (the pore volume given in Table 14), and a value of 2 for the Archies law coefficient. This factor (0.98^2), gives a correction factor of 0.96 for the D, which means that the D in this APE membrane is reduced by ca 4%. By using this equation for the pore volume in Table 14, the following correction factors for D are achieved for the common DGT membranes :0,90 (APA) and 0,71 (CGA). This seem to give reasonable agreements for simple metal ions such as the 5-15% reduction obtained for APA membranes (Garmo, Røyset et al 2013) and for the 20-40% reduction in D obtained for the CGA membrane (Garmo, Røyset et al (2002)), and the ca 30% reported by Zhang and Davison (1999) for the same CGA membrane.

The membrane resistance R can be defined as:

$$R = \frac{D_{DGT}}{D_W} \qquad \text{(Equation 12 in Table 7)}$$

D_{DGT} Diffusion coefficient of the ion in the water saturated membrane matrix

D_W Diffusion coefficients of the ion in water

R may be expressed as an unitless factor from 1 to 0 or in percent from 100 to 0%, as above. For the correction of the D, the R factor must be inverted from 0 to 1, or to 0 to 100%. The resistance corrected diffusion coefficient D_{R_DGT} for a DGT membrane can thus be obtained by the equations

$$D_{R\_DGT} = D_W \; x \; \left(\frac{100-R}{100}\right) \qquad \text{if R in percent from 100 to 0 \%}$$

or

$$D_{R\_DGT} = D_W \; x \; (1 - R) \qquad \text{R as an unitless factor from 1 to 0}$$

The Archie law based pore-volume correction is a step in the right direction, but do probably not correct for the all other parameters influencing the membrane resistance factor especially for organic molecules with larger molecular weights than the common metal cations. Future research needs to improve the understanding of the separate factors controlling the total magnitude of the R factor. The understanding of the basis mechanisms for the membrane resistance will become an important research topic for the DGT community in this new area. To get better understanding of the membrane resistance factors several aspects need to be further explored, such as those dealt with in the studies of transport processes in macroporous media by Logan (2012), and needs knowledge on the influence on factors such as :

- Pores size distributions of the membranes
- Charged sites of the membranes , i.e. a charge density of the membrane material
- Effect of pH. Influences both membrane properties as well as the charge density of the molecules
- Ionic strength effects especially on the membranes
- Molecular weight of the diffusing ions or molecules
- Molecular size and shapes of the diffusing ions and molecules, i.e. molecular diameters or volumes.
- pH and charge of the diffusing ions or molecules
- The frictional factor proposed by Buffle (2014).

3.3.3 Diffusion coefficients (D) and membrane resistance (R) of DOM compounds

We have few data which actually report D and R for DOM compounds in DGT membranes.

Zhang and Davison (1999) studied the diffusion of three humic compounds with Mw from 2400 to 16400 for the most common DGT membranes (see Table 14 for details). The normal open pore agarose polyacrylamide membrane (APA) had rather high penetration of fulvic and humic acids. The so-called constrained APA membrane (CGA) with more narrow pores (pore size was specified to ca 1 nm) was developed to achieve higher exclusion of these compounds. The purpose of the agarose based membrane (AGE) was to develop a membrane with relatively large pore-sizes (>20 nm as specified by Zhang and Davison 1999). The poresize of the AGE membranes are less defined. Lead et al (2003) reports values in the range 30 to 100 nm, while Fatin-Rouge et al (2003) reported that the poresize could be from 1 to 400 nm. Thus AGE membranes are expected to have less restriction on the diffusion of ions and molecules. Since the properties of the AGE membrane may depends on the membrane casting conditions, the production process of the AGE membranes are important to control, in order to produce membranes with specific spore sizes and thereby more accurate separation properties.

Figure 12 shows the D values for these humic compounds in water and in the respective DGT membranes from Zhang and Davison study. The curves for the D achieved from the Buffle2007 equation with R=0 and R=50% are shown. The D values for AMP and IP6 from this work is also shown.

The membrane resistance is calculated by dividing the D of these humic compounds in each membrane with the D in water predicted from Buffles equation (used at no resistance R=0%). Thus R values in each membrane type are achieved. Figure 13 shows these R values plotted versus Mw. The R for AMP and IP6 from this work is also shown (only for the APA membrane).

Table 14
Properties of the most common DGT membranes and the humic substances used for the studies of the diffusion coefficients in the DGT membrane by Zhang and Davison (1999)

Properties of the DGT membranes studied				
Code	Type	Cross-linker %	Pore size	Pore volume *)
AGE	Agarose membrane	No	>20 nm	98
APA	Open pore agarose polyacrylamide membrane	0.3 %	>5 nm (5-10 nm)	95
CGA	Constrained agarose polyacrylamide membrane	0.8%	< 1 nm	84
Properties of the humic compounds studied				
Code	Type	Mw, Da,	Charge meq/g	
AFA	Fulvic acid from water	2400	4.5	
AHA	Humic acid from water	6400	3.5	
PHA	Humic acid extracted from peat	16500	(3.6)	

*) The porevolume is estimated based on the water content of the membrane matrix.
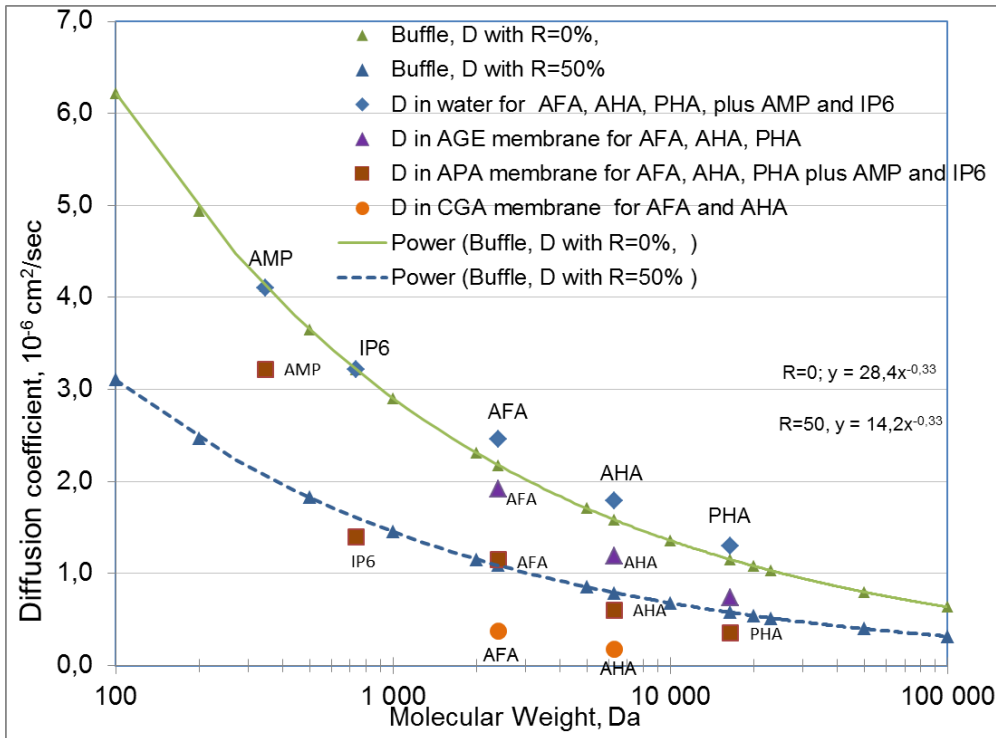
Figure 12.
Diffusion coefficients (D) vs Mw according to Buffle's equation at membrane resistances of R=0 (green) and R=50% (blue). The D values are shown for the three fulvic and humic acids AFA, AHA and PHA (plus AMP and IP6 from this work) in the DGT membranes APA, AGE and CGA (Table 14).

Figure 13.
Membrane resistance (R) vs Mw in the DGT membranes APA, AGE, CGA (see Table 14) for the three fulvic and humic acids AFA, AHA, PHA (and AMP and IP6 from his work) .

The results for D in Figure 12 show the following for the AFA, APA and PHA compounds:
- The diffusion coefficients in water are close to the D-values predicted by Buffle's equation at R=0% i.e. in water.
- In the AGE membrane the D values are in the middle between those for water (R=0) and those at R=50%, i.e at a R between 10 to 30%.
- In the APA membrane the D values are located around Buffles's line for R=50%.
- In the CGA membrane the D is very low and only ca 10% of those in water.

The R values in Figure 13 shows the following for the AFA, APA and PHA compounds:
- The resistance increase with molecular weight for all the 3 membranes. Thus large molecules not only diffuse slower but have higher membrane resistance.
- The membranes with larger pore-sizes show the lowest resistance. I.e. the agarose membrane (AGE) has R in the range 10 to 35%.
- The APA membrane has a R from 20 to 65% for the Mw from ca 1000 to 10000
- The CGA membrane has the highest R of around 80-90%. This membrane has high exclusion of even LMW fulvic acids. This membrane is not appropriate for the collection of DOP compounds. This membrane has also a significant R of 20 to 40% for ordinary metal ions (Zhang and Davison (1999) and Røyset and Garmo (2002)).
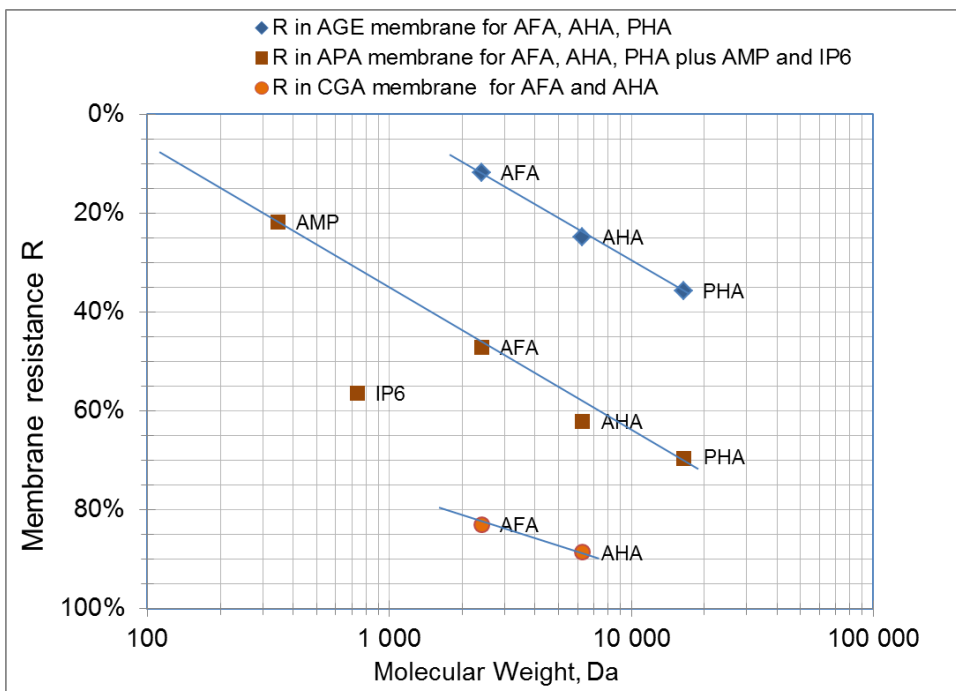
Table 15.

Membrane resistance (R) estimated for different Mw ranges for the APA and AGE membranes according to the data in Figure 13. The Uncertainty of D are calculated as the difference between the average and highest and lowest D value in the Mw range. The uncertainty in percent is calculated by the uncertainty of D divided by the average D in the range.

Molecular weight range	Diffusion coefficient (D) of organic molecules in water according to Buffle's equation (2007)/				Membrane resistance (R)	
	Range of D	Average D in Mw range	Uncertainty of D	Uncertainty of D in %	In APA membrane	In AGE membrane
Da	10^{-6} cm^2/sec	10^{-6} cm^2/sec	10^{-6} cm^2/sec	%	%	%
300 – 10000	4.5 to 1.4	2.9	1.5	50	20 - 65	0 - 35
300 – 3000	4.5 to 2.0	3.3	1.25	38	20 - 45	0 - 15
300 – 1000	4.5 to 3.0	3.7	0.7	20	20 - 35	<5
1000-3000	3.0 to 2.0	2.5	0.5	16	35 - 45	5 - 15
3000 -5000	2.0 to 1.7	1.85	0.2	12	45 - 55	15 - 20
5000-10000	1.7 to 1.4	1.55	0.15	10	55 - 65	20 - 30
10000 -20000	1.4 to 1.1	1.25	0.15	12	65 -70	30 - 35
>20000	<1.1	<1.1	-	-	>70	>35

3.3.4 Can membrane resistance (R) be predicted based on Mw?

The data in Figure 13 can be used as coarse estimates as summarized in Table 15. Although data are preliminary based on few data, the data indicates the following patterns
- In the APA membrane, the data for AFA; AHA and even PHA express an almost linear relationship between R and Log Mw.
- When extrapolating backwards from AFA, AHA and PHA, the R of AMP of ca 20% fits with this R vs log Mw relationship.

- The R of IP6 is very high compared to both AMP and what can be extrapolated from the R vs log Mw relation for AFA, AHA and PHA. This anomalous behaviour compared to the values for AFA, AHA and PHA, is caused by the high charge density of the IP6 molecule (ca 6 negative charges at pH 5). Thus, if the charge density of the molecules increases, the R will move to a larger numerical value than expected from the linear relation drawn through the data points for AFA; AHA and PHA.
- The same linear relationship between R and log Mw is achieved for the AGE (agarose) membrane. The resistance is considerably lower than for the APA membrane, probably due to the larger pore-size of the AGE membrane. The R is below 5 % Mw below 1000 Da. At Mw at 10000 Da the R increases to ca 20%.
- The relatively linear relation between R and log Mw indicates that the R of DOM molecules can be predicted up to Mw between 10000 and 20000 Da. Combined with Buffle's equation some rough predictions of the actual D for dissolved organic compounds containing phosphorus can be obtained. The general pattern is that the R increases with molecular weight. But still at Mw of ca 10000 Da the R factors are not larger than ca 60 % for the APA and ca 30% or AGE membranes.
- Thus, neither the APA nor the AGE membrane seems to have an upper Mw cut off where molecules are completely excluded. This was also found by Pacal et al (2008). Considerable penetration into the membrane was found for particles with diameter over 50 nm. The higher R at larger Mw slows down the overall molecular diffusion movement in the membrane, but there is no clear upper Mw cut off. When the molecular weight specific R factor is obtained, the molecular weight and "membrane specific D" (D_w x R) can be estimated.
- The membrane resistances achieved are summarized in Table 15, divided into in molecular weight ranges from 300 to 20000 Da. We lack some data in the LMW range to get good predictions below 1000 Da especially for the AGE membrane.
- Membrane resistance values versus log Mw have not been published before, and the R values in Table 15 are the best estimates for the time being.
- The uncertainty of the D values is estimated in Table 15. If we only use the average D value of 2.9 to cover the whole Mw range from 300 to 10000 Da, the uncertainty of D is in the order of about 50%. When divided into smaller Mw ranges, the uncertainty of D for each molecular weight range can be reduced to between 10 and 20%.
- The range for the resistance factor is about 10% for each Mw class. The uncertainty in the R term is estimated to be around 5% (half of the range) for each Mw class.

3.3.5 Estimates of diffusion coefficients for the DIP fractions

Li and Gregory's D-values in Table 11 for the 3 predominant orthophosphate species are 8.46, ($H_2PO_4^-$), 7.34 (HPO_4^{2-}) and 6.12 (PO_4^{3-}). These D values are given at the pH where each orthophosphate species has its highest abundance (in middle between each pK_a). The neutral H_3PO_4 dominates at the pH below the pK_{a1} (2.12). The abundance of $H_2PO_4^-$ is 100% at pH 4.67 ((2.12+7.21)/2), HPO_4^{2-} is 100 % abundant at pH 9.94, while PO_4^{3-} starts to dominate (>50%) at pH above 12.67.

Zhang and Davison (1998) reported a diffusion coefficient 6.05 (25 ^oC) for the $H_2PO_4^-$ specie based on a diffusion cell test at a pH of 5 (where the $H_2PO_4^{-s}$ specie dominates). The effect of D at increasing/decreasing pH based on the properties of the different orthophosphate species was not discussed. This D is ca 70 % of the value of 8.46 for $H_2PO_4^-$ in Table 11. The ca 30 % reduction in D compared that of D for the free orthophosphate species in water, was explained by membrane resistance caused by interaction of the negatively charged phosphate species with charged sites in the membrane. Later DGT works (Panther et al., 2010 and Teasdale et al., 2013) have used the D value of 6.05, without reporting or questioning the influence of pH on the D for the orthophosphate species.

Table 16 shows this pH dependence of the D. The D values were calculated at each pK_a value (where the distribution between the 2 ion pairs are 50-50) and at the pH in the middle between the pK_a where each species have its maximum abundance (100%). The corresponding D values at R=30 % are also used as proposed by Zhang and Davison (1998).

The data from Table 16 are shown graphically in Figure 14 in the pH range 4 to 13 for both the original D values from Li and Gregory (1974) (R=0%) and those with R=30%. In the pH range from 4 to 9 (which is expected in natural water) the D value decline from about 8.5 to 7.3 at R=0 and from 5.9 to 5.1 at R=30%. The slopes of the curves in Figure 14 shows that the D value decline per pH unit with a factor of 0.21 (R=0 %) and 0.15 (R=30%).

Davison and Zhang's (1998) D value of $6.05 \cdot 10^{-6}$ cm^2/sec (R=30%) agrees better with the D values by Li and Gregory, and also with the D values of $6.0 \cdot 10^{-6}$ cm^2/sec at pH 5.1 and $5.8 \cdot 10^{-6}$ cm^2/sec at pH 6.3 obtained by Røyset et al. (2004). Although the uncertainty of the D value in this latter dataset is ca 0.5, the change of 0.2 D units by 1.2 pH units from pH 5.1 to 6.3 is close to the change in D of 0.15 per pH unit obtained by equation in Figure 14.

According to these results the calculations in Figure 14 and Table 16 seem to be sound. The D values of the orthophosphate species need be adjusted for the effect of pH. This pH dependence has not been described earlier. This is a new functionality for the orthophosphate species. These equations depend on the properties of membrane, and have to be re-evaluated for other membrane types. Based on the results in Figure 14, two correction equations are achieved, one at R= 0% (for water) and one at R=30% (for the normal agarose polyacrylamide membrane (APA)):

$D = -0.15 \cdot pH + 6.62$ (R = 30% for the APA membrane)   (Eq. 13 in Table 7))
$D = -0.21 \cdot pH + 9.46$ (R= 0%, i.e, free diffusion in water)

If this correction is not performed an uncertainty of D from 0.15 to 0.20 per pH unit is assumed. This uncertainty of the D is then from 3 to 4 % if the D value is not adjusted for the nearest pH unit (uncertainty based on 0.2 divided by D of 6). A D of $6.0 \cdot 10^{-6}$ cm^2/sec was chosen as a default value. This pH dependence is further studied in Chapter 4.5.

Table 16

Diffusion coefficients (D) for the different orthophosphate species, shown at the pH in the middle between two pK$_a$'s (where the individual species dominate), and at each pK$_a$ (where the distribution of the two species is 50%).

Ortho phosphate species	Abun- dance, %	pK$_a$	pH	D by Li & Gregory (1974)	
				In water at R=0%	Membrane resistance 30%
				10^{-6} cm^2/sec	10^{-6} cm^2/sec
H$_3$PO$_4$	>50		<2.12		
H$_3$PO$_4$ / H$_2$PO$_4^-$	50 - 50	pKa$_1$ 2.12	2.12		
H$_2$PO$_4^-$	100		4.47*	8.46	5.92
H$_2$PO$_4^-$ / HPO$_4^{2-}$	50 - 50	pKa$_2$ 7.21	7.21	7.90**	5.53**
HPO$_4^{2-}$	100		9.94*	7.34	5.14
HPO$_4^{2-}$ / PO$_4^{3-}$	50 - 50	pKa$_3$ 12.67	12.67	6.73**	4.71**
PO$_4^{3-}$	>50		>12.67	6.12	4.28

* pH calculated where each specie has maximum (100%) abundance, i.e. as the average of the pK$_a$ above and below (pH$_{m1,2}$ = (pK$_{a1}$ + pK$_{a2}$)/2) and (pH$_{m1,2}$ = (pK$_{a1}$ + pK$_{a2}$)/2.
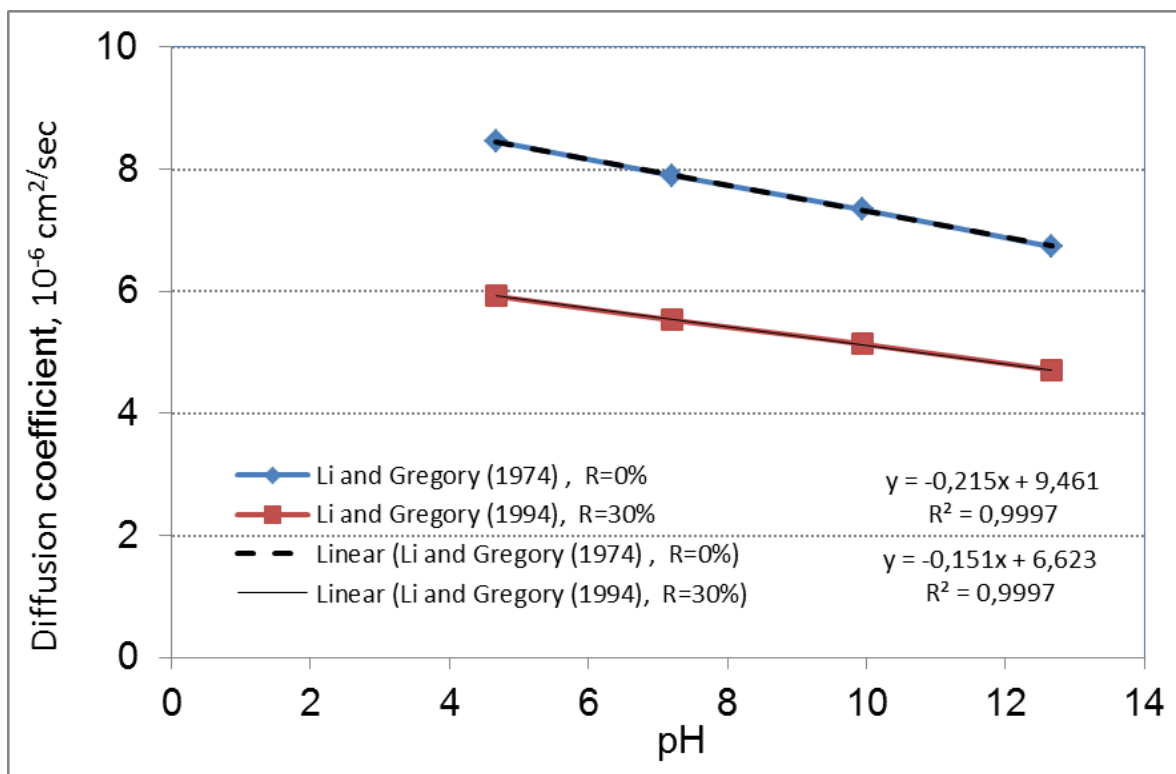**D calculated at the pH of pK$_{a1}$ and pK$_{a2}$.

Figure 14.
The influence of pH on the diffusion coefficient for the orthophosphate species in Table 15, according to the original data by Li and Gregory (1994) calculated without and with 30 % membrane resistance (R).

3.3.6 Estimates of diffusion coefficients for the DOP fractions

Evaluation of the result for AMP and IP6

The experimental data for the D value obtained for AMP of ca 3.16 (±0,44) is about 20 % lower than the D of $4.0 \cdot (10^{-6} \, cm^2/sec)$ calculated from Buffle's equation (Table 10). This deviation is probably mainly caused by resistance through the diffusion membrane, as the AMP molecule carry ca 2.5 negative charges according to its 3 pK_a values (0.9, 3.4 and 6.1, Figure 6), at the pH between 5.0 and 5.5 of these experiments.

The experimental D value for IP6 of $1.3 \cdot (10^{-6} \, cm^2/sec)$ is ca 40% of $3.2 \cdot (10^{-6} \, cm^2/sec)$ calculated by Buffle's equation, and implies a large membrane resistance. The 12 pK_a values for the protons on the 6 phosphate groups attached to IP6 lie in the range 1 to 12, with 5 below 5.5 and further 2 below 6.8 (see Figure 6). At pH of 5.5 (used in this laboratory experiment) at least 5 of these phosphate groups are fully ionized and IP6 carry thus a negative charge of at least 5. The charge density of the small IP6 molecule is thus high and causes the strong resistance of ca 60 % (ca 40 % of the calculated D).

The R of AMP of ca 20 % fits with the extrapolations in the resistance obtained from the humic compounds in Figure 13. IP6 has a strong negative deviation from this R curve due to the strong negative charge and high charge density.

D values for the field samples in this work

The literature contains few data on the diffusion coefficients of typical dissolved organic phosphorus compounds in water. The best source is data from DOM compounds in water, as the molecules of the DOP fraction may be considered to have similar properties as DOM molecules containing phosphorus groups. The dissolved organic compounds in the DOP fraction are expected to be present in natural water at molecular weights from less than 300 Da to at least 10 000 Da (Worsfold et al 2008., Ged and Boyer, 2013)). We can expect a r mixture of both low molecular and high molecular weight DOP compounds. Buffle's equation is useful to predict D for the individual DOP compounds groups as soon as the Mw distributions of these are known or estimated. The R factor established in chapter 4-3.4 introduces a new tool to adjust the D based on properties of the membranes.

The molecular weight distribution is not available for the DOM/DOP molecules collected by the DGTs in the field study at Lake Vansjø. Some assumptions for the Mw distributions are needed to predict the D and R value. In natural water draining forest soils the Mw range of DOM/DOP molecules may be from ca 300 to at least 10000 Da. Ged and Boyer (2013) found considerable Mw fractions of DOM in the range 5000-10000 Da in runoff from the Everglades area. The Mw of DOP molecules in runoff from agricultural soils are assumed to be in the LMW to MMW range (Turner et al 2003), with a larger proportion probably in the LMW range <1000Da. If the predominant Mw range is from 300 to 3000 Da, a D-value around $3.0 \cdot (10^{-6} \, cm^2/sec)$ may be a first approximation for the D for the DOP fraction for the agricultural water runoff samples in this study.

The pH dependence of D for the humic compounds

The D of the orthophosphate species decreases with increasing pH due to the increasing negative charge of the orthophosphate molecule. DOM compounds are poly-protic organic compounds where the negative charge increases with pH due to deprotonation of functional groups. Do the D of the DOM compounds also decrease as the molecular charge increase with higher pH. ? Lead et al. (2003) studied very detailed the effect of pH on D for some fulvic and humic acids. The D increased with ca 0.5 numerical units from pH 3 to pH 7, i.e.an increase of ca 0.1 per pH unit. This is the opposite direction of change compared to the orthophosphate species. Lead suggested that the humic substances form molecular aggregates (2 or more molecular units) with larger hydrodynamic diameter at lower pH. The larger hydrodynamic diameter of these molecular aggregates slows down the diffusion rates, and the resistance in the membrane increases. These aggregates disperse

when pH increases, the hydrodynamic diameter become lower and the D is expected to increase. There is no data for how the D behaves at pH above 8. Above this pH the humic compounds to become more negatively charged and the D is expected to decrease as for the orthophosphate species.

3.4 Sampling precision in the field

During the field study 4 samplers of Fe-DGT and Me-DGTs were deployed in parallel. The sampling precisions achieved are given in Table 17. For the Me-DGT the sampler number 4 was damaged and omitted from the calculations. The sampling precision (RSD) varied from 6 to 20%, for both Fe-DGTs and Me-DGTs for all the three DGT fractions (DTP, DIP and DOP). The RSD is lowest for the DGT-TDP fraction (6-9 %) and highest for the DGT-DOP fraction (18-19 %). This was as expected as the DGT-DOP fraction has the lowest concentration. However, the absolute standard deviations in concentration units are in the range 1.5 to 4.1 µg P/L for all the three fractions. This is in the same range as achieved in the lab experiments estimating the D, and is thus considered acceptable.

Table 17.
Sampling precision of P fractions in the field for Fe-DGT and M-DGT samplers deployed in the Støa stream June to September 2011 (Parekh, 2012). The Me-DGT sampler parallel 4 was damaged during deployment, and the data for this parallel was omitted in the calculations.

	Ferrihydrite Fe-DGT			Metsorb™ Me-DGT		
	DGT-DIP	DGT-DOP	DGT-TDP	DGT-DIP	DGT-DOP	DGT-DTP
Parallel	µg P/L	µg P/L	µg P/L	µg P/L	µg P/L	µg P/L
1	24.3	7,1	31.4	32.5	10.5	43.0
2	32.6	5.8	38.5	31.4	14.0	45.4
3	27.7	9.1	36.7	24.9	15.1	40.0
4	25.9	8.7	34.5	na	na	na
N	4	4	4	3	3	3
Average	27.6	7.7	35.3	29.6	13.2	42.8
Std. dev.	3.6	1.5	3.0	4.1	2.4	2.7
RSD %	13.0	19.4	8.6	13.9	18.1	6.4

3.5 Comparisons of uptake on Fe-DGT and Me-DGTs

Panther et al. (2010) reported that Metsorb™ has higher adsorption capacity (ca 37 µg compared to ca 7 µg of P) and higher selectivity for orthophosphate than Ferrihydrite. The higher capacity for Metsorb™, permits higher uptake especially at higher ionic strengths (such as in sea water), and also longer deployments times may be possible. During the field study the Me- and Fe-DGTs were deployed in parallel and the results are shown in Figure 15.

The uptake of the DGT-TDP and DGT-DIP fractions on the Me-DGT fraction were found to be slightly higher than for the Fe-DGT, as the slopes of their regression correlation are 0.95 and 0.97, respectively. For the DGT-DOP fraction the uptake on Me-DGT seems to be higher as the slope of the Fe- vs Me-DGT correlation curve is 0.89.

In general the differences are not large, especially considering the uncertainties addressed above. This indicates that both samplers behave similarly to each other in these surface water samples both for the DGT-DIP and DGT-DOP fraction. Both the ionic strength and the concentration of P

compounds are relatively low in these surface waters. An overloading of capacity is therefore not expected within the deployment period used in this study. The capacity benefit of Metsorb™ is not visible in these data.

Both samplers seem to have practically equal performance in these surface waters. The good performance of Fe-DGT is promising. Fe-DGT is a more convenient sampler for laboratory work due to the quantitative extraction only by sulphuric acid. The DGT extract can be directly analysed by MBM. In a very recent study of Price, Teasdale et al. (2013), they concluded that the adsorption of Fe-DGT was as high as for the Me-DGT. Thus, making adsorbent with high capacity will thus be an important research issue. Is the low capacity of the Fe-DGT adsorbent caused by that this adsorbent layer has not been produced correctly to give higher capacity?. This topic needs to be addressed in future research, as ferrihydrite is easier to handle than the Metsorb™ adsorbent.

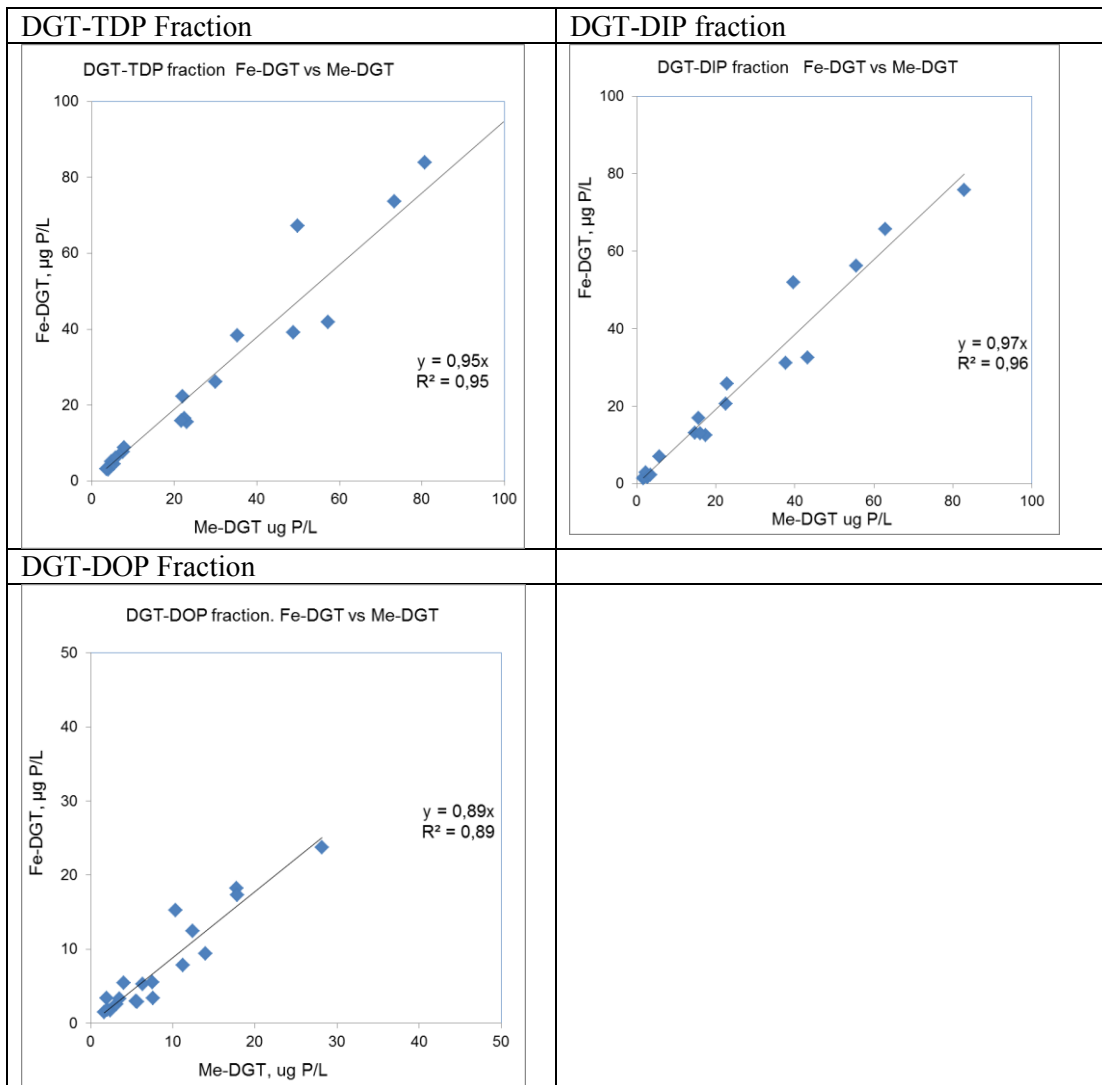| DGT-TDP Fraction	DGT-DIP fraction

DGT-DOP Fraction	

Figure 15.
Uptake by Me-DGT compared to Fe-DGTs for the three fractions DGT-DTP, DGT-DIP and DGT-DOP. Data from the field campaign June to September 2011. The slopes in the graphs are independent of the values of D, as the same D value is used for both the Fe- and Me-DGT samplers.

3.6 Evaluation of the data from the field sampling

The DGTs were deployed from 6 to 14 days. The concentrations of the DGT fraction were compared to the corresponding stream water phosphorous concentrations. The intention was to collect grab water samples at the start and stop of each deployment period. For practical reasons this was not always possible to accomplish. Where grab samples at start and stop were missing the DGT concentrations had to be compared to the stream sample collected either at start or stop of the DGT deployment period. A recalculation was made where all the collected grab sample data were averaged between the start and stop of the DGT deployment periods. The possible deviation between the two approaches was tested by slopes of the DGT vs grab sample data (as shown in Figure 16). The change in the slope by this recalculation was less than 5 %, and the original data were therefore used.

The comparisons where first performed with the initially chosen diffusion coefficients of 6.0 (DIP) and 3.0 (10^{-6} cm^2/sec) (DOP), based on the initial assumptions made in Chapter 4.3. These result are shown in Figure 16 (scatter plots) and in Figure 17 (bar plots).

3.6.1 The relation between D, sampling rate and calculated concentration

The passive sampler equation has an implication which is very important to be aware of. Higher D values give lower time averaged concentrations. The D value is the "hearth "of all DGT calculations. The influence of the diffusion coefficient is obvious when examining the DGT equation 3 in Table 7:

$$DGT \; average \; concentration = m / [t \, (DA/L]$$

According to the uptake rate term (DA/L) the sampled amount per time unit increases with D. An intuitive impression is that higher diffusion rates (D) gives higher calculated concentration. However, since the diffusion coefficient (in the uptake rate (DA/L) is in the denominator of the DGT equation, the effect of D on the calculated average concentration is the opposite. This relationship applies to DGTs and passive sampling in general. At a fixed sample time, this relation implies the following:

- Increase in D give higher DGT sampled volume, and thus lower calculated average concentration.
- Decrease in D gives lower DGT-sampled volume, and thus higher calculated average concentration.

At D-values of 6.0, 3.0 and 1.5 (10^{-6} cm^2/sec), the DGT-sampling rate decrease from ca 16, 8 and 4 mL/day, respectively. When the D is not known such as for the DOP fraction in this work, changing the D gives large influence on the calculated average concentration. This is the basis for the modelling performed below in order to find the D value where the DGT fractions which fits best to the corresponding stream water fractions.

The slopes for the linear regression for both the DGT-DIP and DGT-DOP fractions using these initial D values showed deviations from unity. The effect of D can be examined by recalculating the result in Figure 16 by altering the D values, and display the result in similar way. The new concentrations for the DGT fractions can then be compared with the corresponding water fractions, to investigate which value of D give the best fit to the stream water data. This was done and the results are shown in Figure 18 (DIP) and 19 (DOP). The data from the linear regression and correlation coefficients are shown in Table 18 to give a full overview of the linear regressions equations achieved.

Table 18.
Overview of linear regressions equations (y =a x) and correlation coefficients achieved from the data shown in Figure 16, 17 and 18. The DGT-TDP is calculated as the sum of DGT-DIP and DGT-DOP.

	Water	DGT	D (10^{-6})	Range	Fe-DGT	Fe-DGT	Me-DGT	Me-DGT
	Y	X	cm^2/sec	µg P/L	Slope (a)	R^2	Slope (a)	R^2
Figure 16	TDP	DGT-TDP	Sum *)	1 - 125	1.039	0.66	1.022	0.758
	DIP	DGT-DIP	6.0	1 - 110	1.10	0.757	1.094	0.665
	DOP	DGT-DOP	3.0	1 - 28	0.856	0.173	0.768	0.218
Figure 18	DIP	DGT-DIP	6.0	1 - 110	1.102	0.757	1.094	0.665
	DIP	DGT-DIP	5.5	1 - 110	1.010	0.757	1.003	0.665
	DIP	DGT-DIP	5.0	1 - 110	0.918	0.757	0.912	0.665
Figure 19	DOP	DGT-DOP	4.0	1 - 28	1.141	0.173	1.024	0.216
	DOP	DGT-DOP	3.7	1 - 28	1.055	0.173	0.947	0.216
	DOP	DGT-DOP	3.0	1 - 28	0.856	0.173	0.768	0.216
	DOP	DGT-DOP	2.0	1 - 28	0.571	0.173	0.512	0.216

*) The DGT-TDP fraction is the sum of the DGT-DIP and DGT-DOP fraction with the D of 6.0 and 3.0 (10^{-6} cm^2/sec), respectively.

3.6.2 The DGT-TDP fraction

For the TDP fractions the slopes of TDP vs DGT-TDP, are close to 1.0 (1.022 and 1.039), for the Fe-DGT and Me-DGT, respectively. The correlation coefficients are reasonably good (0.66 and 0.0758).

The DGT-TDP fraction is a sum parameter (sum of DGT-DIP and DGT-DOP, see Chapter 3.3). Thus the DGT-TDP fraction includes an uncertainty contribution from both the DGT-DIP and DGT-DOP fractions. The slopes above are close to 1.0, but the uncertainties above makes it difficult to draw clear conclusions of the agreement between DGT-TDP vs the stream water TDP fraction. For this purpose the results from the DIP fractions is expected to be better, as this fraction is based on a direct measurement of the orthophosphate fraction in the extracts from the DGT.

3.6.3 The DGT-DIP fraction

The diffusion coefficient of 6.0 (10^{-6} cm^2/sec) used for the orthophosphate species of DIP fractions has earlier been assumed to be reasonably accurate (chapter 4.2). The adsorption of the ortho-phosphate species is expected to be close to 100% on both adsorbents (Zhang et al. 1998, Mason et al. 2008). The D coefficient for the orthophosphate species decreases with ca 0.15 per pH unit (Figure 12, equation 13 in Table 7). The expectation is that the DGT-DIP fraction should be comparable to the water DIP-fraction, but we need to examine if the agreement can be improved by adjusting the D for pH. According to Parekh (2012) the median (upper and lower quartiles) of pH in the stream water samples are: from Støa 7.2 (6.8 to 7.5), from Huggenes 7.2 (6.5 to 7.3) and from Dalen 4.4 (4.2 to 4.6).

Using the initial value of D of 6.0 ($10^{-6}cm^2$/sec), the linear regression slopes (a) of the DIP vs DGT-DIP fractions are around 1.1 (1.10 and 1.094)) for the Fe-DGT and Me-DGT respectively. The correlation coefficients (R^2) are high (0.757 and 0.665). The slope of ca 1.1 indicates ca 10% deviation from the correct D value (Figure 16).

Figure 18 shows results for the DGT-DIP-fractions the recalculated with the three D values 6.0, 5.5 and 5.0·(10^{-6} cm^2/sec). The best fit between the DGT-DIP and stream water DIP is achieved with a D-value of 5.5, where the slopes are very close to 1.0 (1.00 and 1.01 respectively).

For the forest runoff water (Dalen) the pH are between 4.2 and 4.6. The expected D between pH of 4.2 to 4.6 should be between 5.99 and 5.93 (10^{-6} cm^2/sec) according to equation 13. However, the stream water DIP fraction at Dalen is only 1 to 2 μg P/L, and it is not possible to make any meaningful correlation in this low concentration range.

The concentration range for the DIP fraction is dominated by the Støa and Huggenes stations with river DIP from 10 to 110 μgP/L and with a pH range 6.5 to 7.5. The linear regression coefficients of Table 18 for DGT-DIP vs water DIP are thus dominated by the data for the Støa and Huggenes samples with pHs from 6.5 to 7.5.

Based on equation 13 the calculated D at pH of 6.5, 7.2 to 7.5 are 5.64, 5.54 and 5.49 respectively. At the average pH of 7.2 the calculated D is 5.54. This good fit achieved with the D adjusted to 5.5·(10^{-6} cm^2/sec), confirm that the pH correction function for the ortho-phosphate developed in this work improves the accuracy of the calculated DGT-DIP concentrations. The pH effect contributes to ca 0.15 numerical units of the D per pH unit.

3.6.4 DGT-DOP fractions

For the DOP fractions the linear regression slopes of DGT-DOP (at a D of 3.0) vs stream water DOP is 0.856 and 0.768 for the Fe-DGT and Me-DGT, respectively (Figure 16). The linear regression slopes for DGT-DOP vs stream water DOP deviate about 10% for the results from the Fe- and Me-DGT sampler (0.856 and 0.768 above at D of 3.0).

The correlation coefficients of ca 0.2 are poor (0.173 and 0.218). This low correlation for the DOP fractions may have several reasons: The short concentration range (1 to 28 μgP/L), give poor correlation coefficients, although the scatter of the data points is about the same magnitude as for the DIP fractions (Figure 16 D). The lower slopes of ca 0.8 between the DGT-DOP and stream DOP fractions are most likely caused by the uncertainty of the diffusion coefficients. The diffusion coefficients for the DOP fractions influence the calculated DGT-concentrations, and thereby also the slopes above (see chapter 4.2).

To get better insights on the influence of the D for the fit between DGT-DOP and stream water DOP-fractions, the data in Figure 16 was recalculated using four values for D of 4.0, 3.7, 3.0 and 2.0 (10^{-6} cm^2/sec). The results in Figure 19 shows the best fit at the fairly high D-value of 3.7 10^{-6} cm^2/sec (slopes of 1.05 and 0.95). The correlation between the DGT-DOP and the stream water DOP fractions based on the suggested D value of 3.0 (chapter 4.29. gave linear regression slope coefficients 0.76 and 0.85. For the low D value of 2.0 10^{-6} cm^2/sec, the calculated DGT-DOP fractions are about 50 % higher than the stream water DOP fraction (slopes 0.57 and 0.51).

For the forest runoff data (Dalen) the concentrations are below 5 μg P/L.(Figure 17). It is therefore not possible to test the assumption that a lower diffusion coefficient (due to larger molecular weight) would give better fit to the forest stream water DOP fraction. The correlation above in Figure 19 are mostly based on the data from the DOP fractions from the sites Støa and Huggenes, where the DOP fractions are from 5 to ca 25 μg P/L.

Based on these calculations the best D value for the DOP fraction seem to be around 3.7·10^{-6} cm^2/sec. According to Buffle's'equation, this high D value implies that the molecular weights of the DOP fraction collected by the DGT in the agricultural runoff are around 500 to 1000 Da.

DOM compounds in water are expected to contain a significant fraction with Mw in the range 1000 -10000 Da. Ged and Boyer (2013) reported up to 30% of the DOP fraction with Mw above 10000 Da in the Everglades areas (Florida). In the reviews by Turner et al. (2003), it is stated that the DOP compounds in agricultural soils consists of predominantly LMW fractions below 1000 Da. In a recent study in lake sediments, 20 to 50% of the organic phosphorus fractions consisted of mainly LMW "phytate like" compounds (Zhu et al. 2013). The first intuitive impression was that a dominating Mw range from 500 to 1000 is too low. To investigate this further some simplified modelling was done based on the DGT equation. If only parts of the DOP compounds in the stream water DOP fraction is collected by the DGT, we can adjust the collected amount by a Collection efficiency factor:

C_{Ef}: Collection efficiency factor

The C_{EF} factor correct for not quantitative adsorption at the DOP molecules at the adsorbent surface, meaning that the molecules diffuse through the membrane but are not adsorbed. The net effect is that the sample amount m should have been higher.

If the molecules pass the membrane, but the free diffusion is hindered by membrane resistance, then the D must be corrected by the R factor, as explained in chapter 4.3-

Both these correction factors can be put into the DGT equation for further modelling:

$$Co = \frac{m/C_{EF}}{t \cdot (D \cdot R \cdot A/L)}$$

Based on this general model the data set from Figure 16 were recalculated based on the following assumptions:

The stream water DOP fractions are not changed. We assume that the DOP fraction in the stream water is correct, or little affected by the choice of filters and other operational conditions of the water P fractionation method. These assumptions are probably valid, as the uncertainties of the D for the DGT-DOP fraction is much larger than the uncertainties of the method for the water DOP fractions.

1. Keep the amount of DOP the same (i.e the *m* term) , but multiply *D* with the R_T factor from 100 % to 50%. This has the same effect as decreasing the numerical value of D.
2. Adjust the collected amount (*m*) of the DOP fraction by dividing by the C_{EF} factor from 1.0 to 0.5. This is based on the assumption that the DGT only collect parts of the DOP fraction in the water, and that the DOP fraction in water is actually higher than what can determined by the DGT sampler. (i.e. the collected amount *m* on the adsorbent should be higher).

In both cases the result achieved for the recalculation for the DGT-DOP fraction change in the same direction. The DGT-DOP fraction becomes higher than the water DOP fractions by both approaches.

Thus it is not possible to make any clear conclusion on the assumption that part of the DOP fraction is not collected by the adsorbent. It is not possible to conclude that the DOP molecules face a high membrane resistance. These best fit with the high D value of 3.7 (10^{-6} cm^2/sec) indicates that the Mw of the DOP fraction in the runoff water from the agricultural sites are low molecular with Mw in the range 300 to 3000 Da, and that the membrane resistance of these compounds are R is low.

At present the data does not contain information which can explain these deviations any further, and more research is needed (chapter 5.2).

Nevertheless, the comparison of the DGT-DOP and stream water DOP fractions provides some rough approximation of the diffusion coefficients. It is interesting to note that such calculations also can give an indication of the range for the molecular weights of the molecules in the DOP fraction collected by the DGT. Moreover, the models based on Fick's law, the DGT equations in Table 7, the formula based on Buffle's equation, as well as the new knowledge on membrane resistance vs Mw developed in this work, are useful new knowledge which can be further developed.
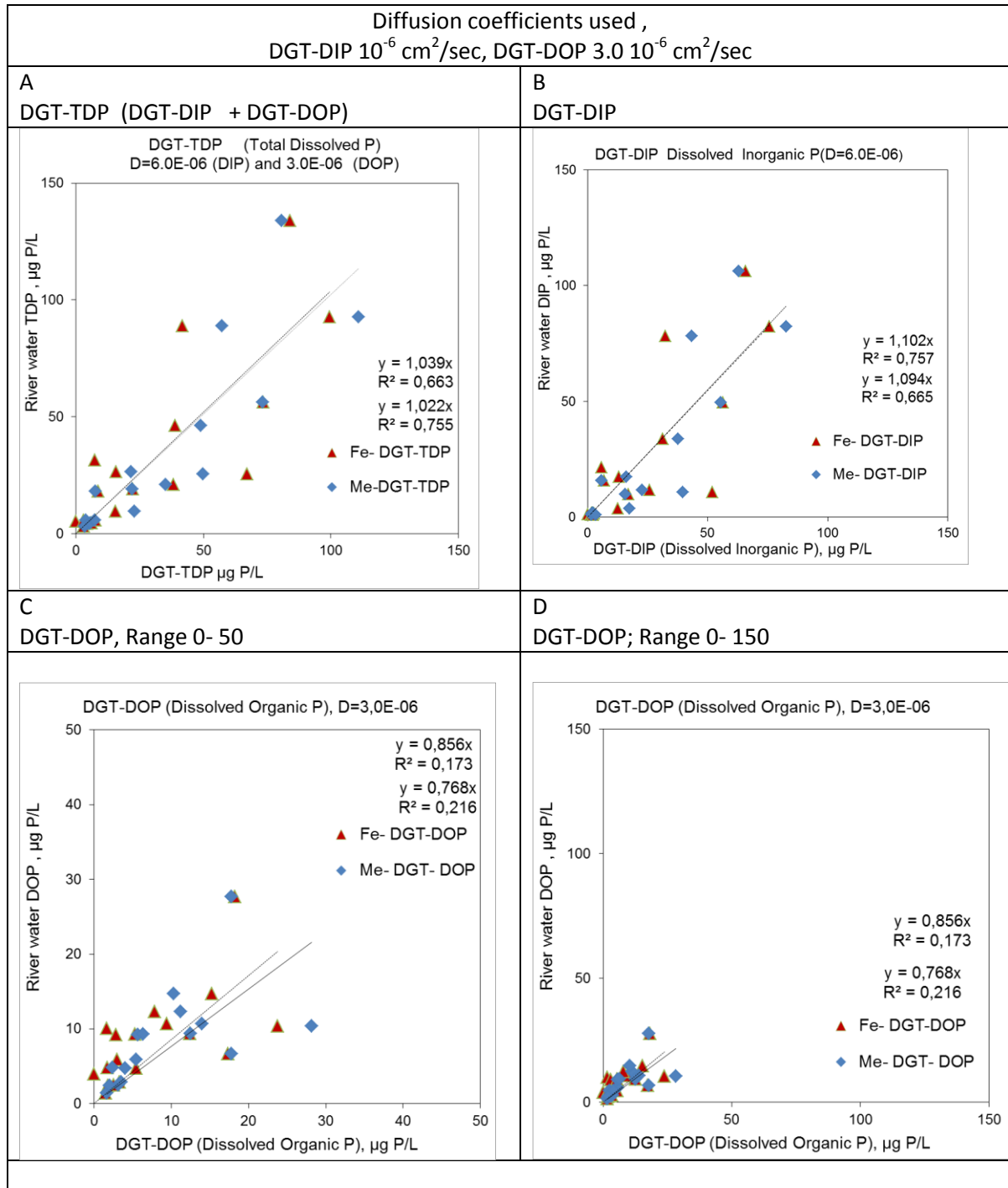
Figure 16.
TDP, DIP and DOP-fractions in stream water compared with the corresponding Fe-DGT and Me-DGT fractions in all 3 streams (Dalen, Støa and Huggenes) (Field campaign June- September 2011). The DGT-DOP fraction is shown in two ranges (C and D) to visually compare the scatter with the TDP and DIP fractions (graph A and B, with D). The data are shown with the initial chosen diffusion coefficients of $6.0 \cdot 10^{-6}$ cm^2/sec for DIP, and $3.0 \cdot 10^{-6}$ cm^2/sec for DOP.
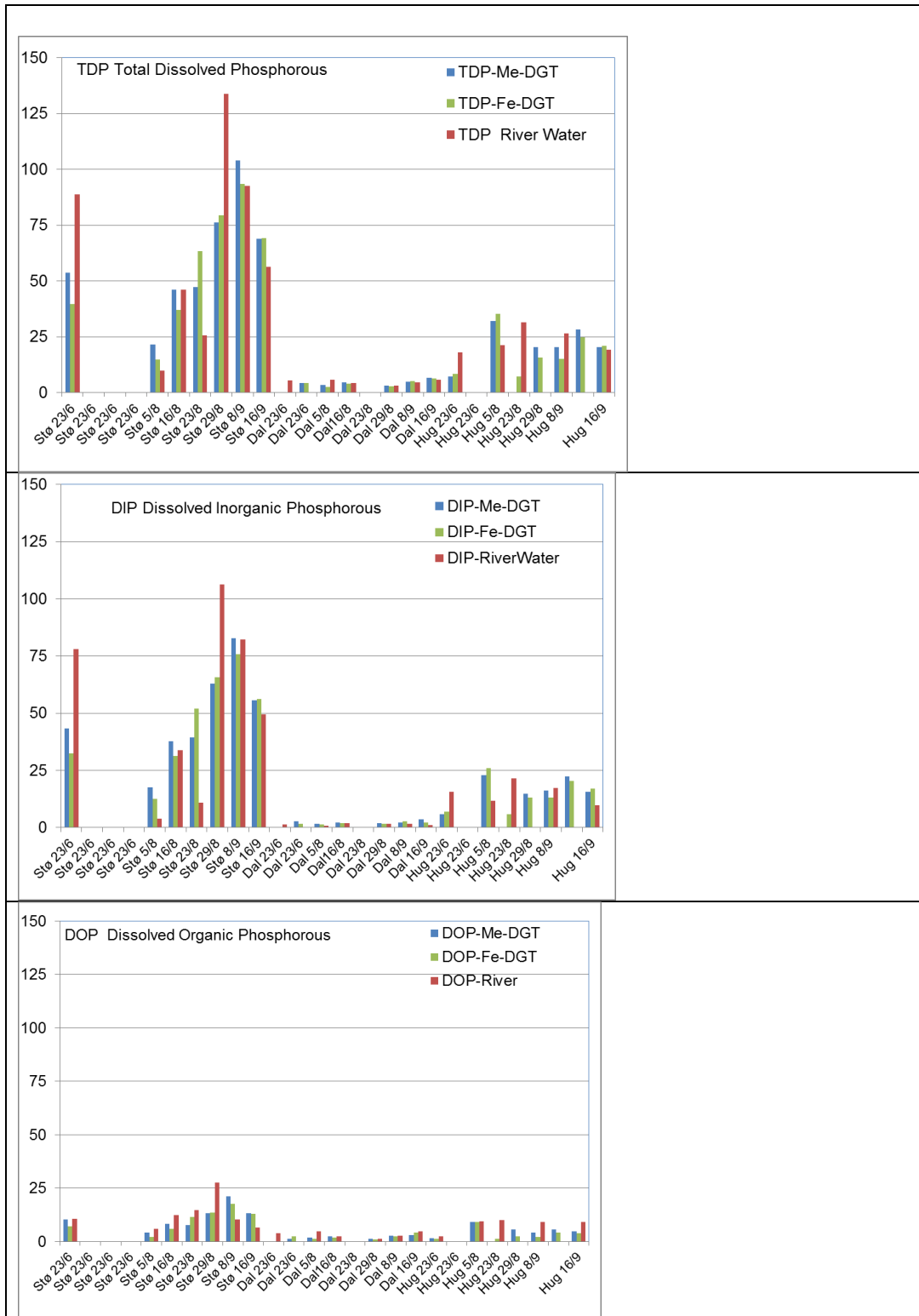
Figure 17.
TDP, DIP and DOP-fractions in stream water compared with the corresponding Fe-DGT and Me-DGT fractions at the 3 stations with agricultural runoff (Støa (Stø), mixed runoff (Huggenes (Hug) and forest runoff (Dalen (Dal)) (Field campaign June- September 2011). The data are shown with the initial chosen diffusion coefficients of 6.0 10^{-6} cm^2/sec for the DIP, and 3.0 10^{-6} cm^2/sec for the DOP- fractions.

DGT-DIP fractions achieved at different values of the Diffusion coefficient (D) as shown in each graph A to C	
A D=6.0 10^{-6} cm^2/sec	B D=5.5 10^{-6} cm^2/sec
C D=5.0E-06 cm^2/sec	

Figure 18
DIP -fractions in stream water compared with the corresponding Fe-DGT and Me-DGT fractions
the 3 stations in all three streams (Field campaign June- September 2011).The DGT-DIP fractions
are calculated with different diffusion coefficients (D) as shown in each graph.

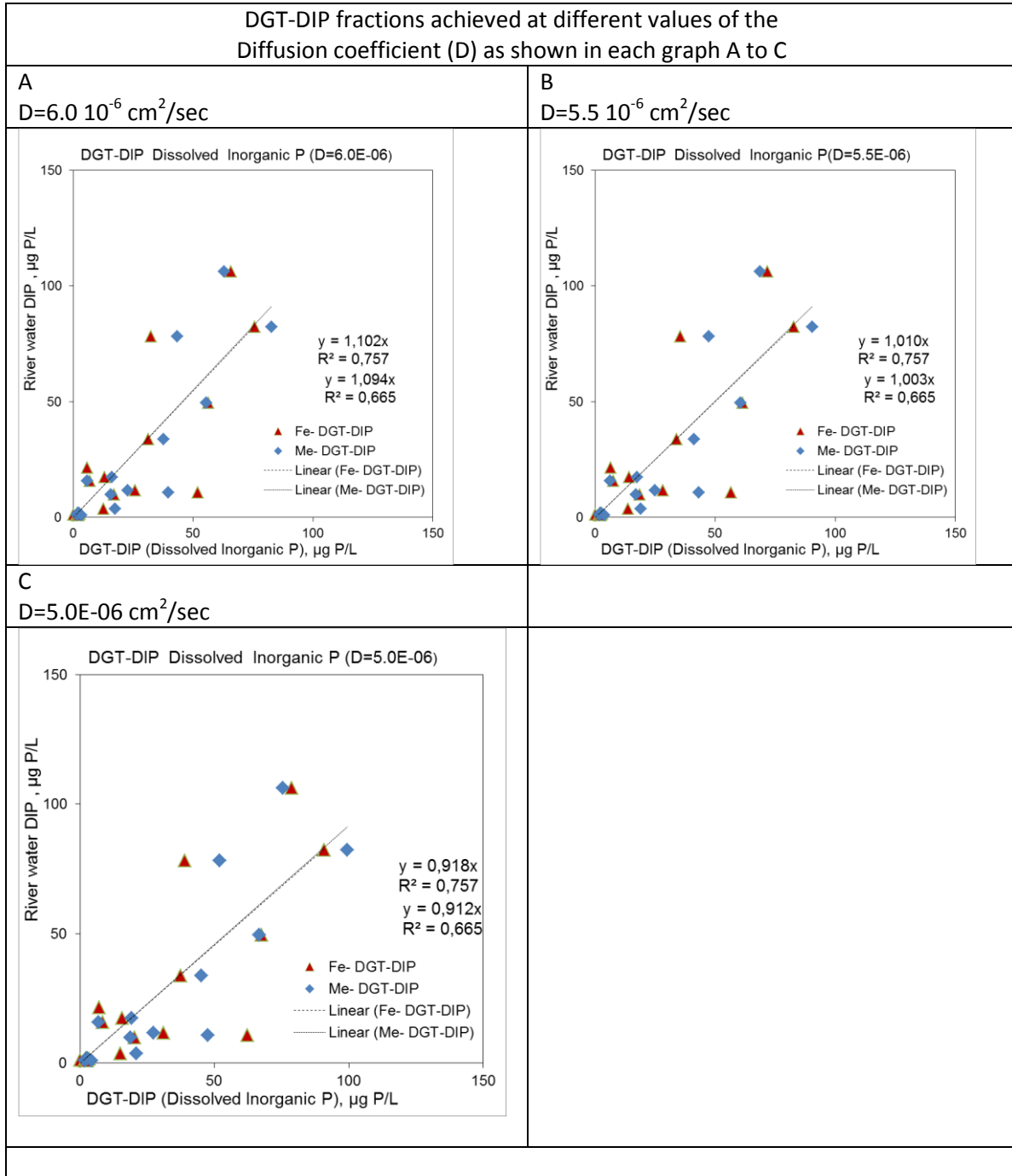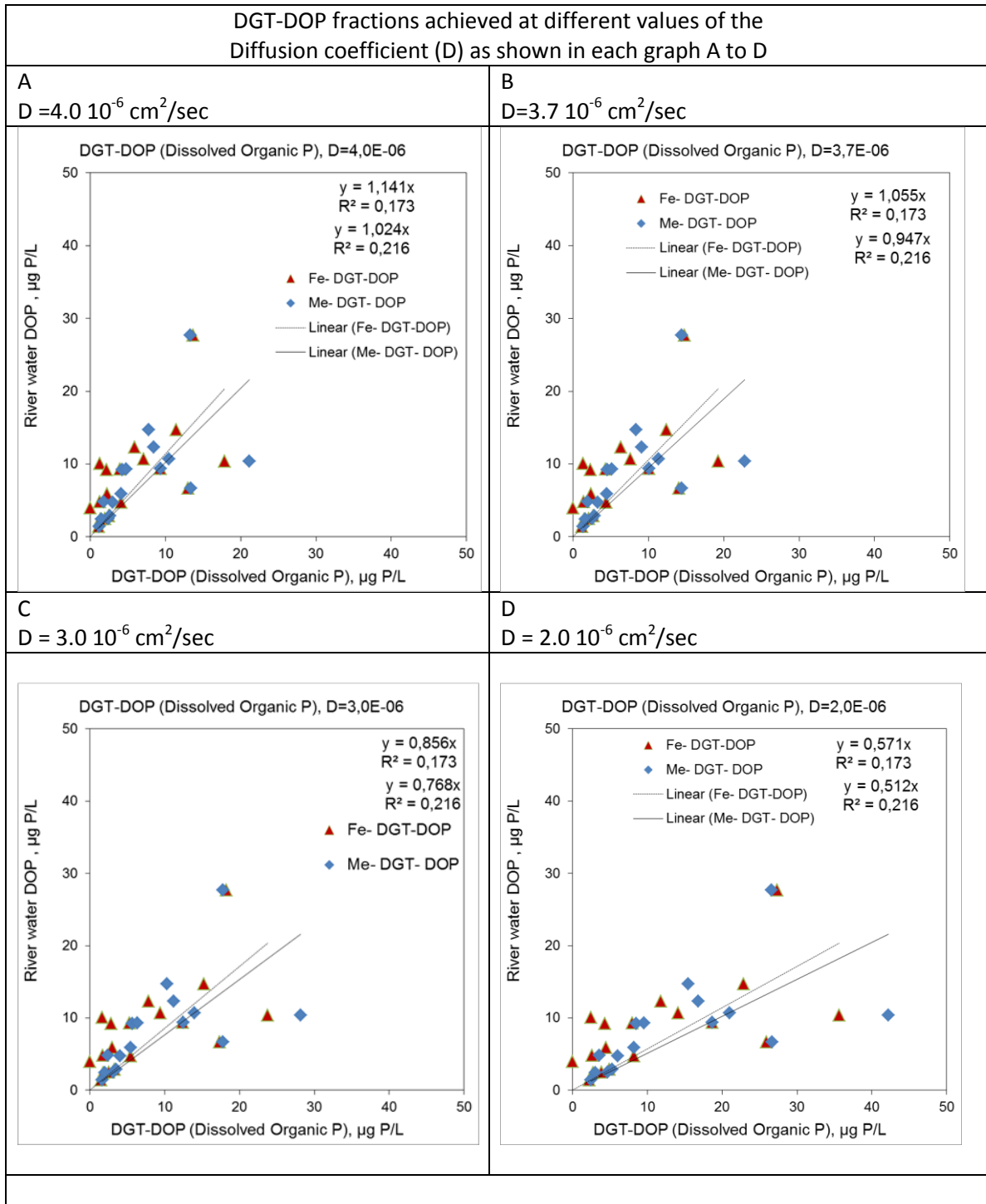| DGT-DOP fractions achieved at different values of the Diffusion coefficient (D) as shown in each graph A to D	
A D =4.0 10^{-6} cm^2/sec	B D=3.7 10^{-6} cm^2/sec
C D = 3.0 10^{-6} cm^2/sec	D D = 2.0 10^{-6} cm^2/sec

Figure 19
DOP-fractions in stream water compared with the corresponding Fe-DGT and Me-DGT fractions in all three streams (Field campaign June- September 2011).The DGT-DOP fractions are calculated with different diffusion coefficients (D) as shown in the each graph.

3.7 Properties of the DOP compounds in the water from the Morsa catchment

3.7.1 DOP/TDP ratios at the 3 sites

Figure 20 shows the DGT-DOP and DGT-TDP-fractions from the 3 sites examined: Støa (mainly agricultural land use), Huggenes (mixed agriculture and forest land use) and Dalen (mainly forest area in the north of Vansjø). Two source patterns are identified. In samples from the predominantly agricultural influenced streams Støa and Huggenes, the DOP/TDP ratio is in the range 20 to 30%. The DOP/TDP ratio is about the double (40 to 60%) in the DGTs deployed in the Dalen stream with DOM rich water from acid forest soils. According to the data in Table 20 the DOC at Dalen is ca 3 times higher (20 to 40 mg C/L) than at Støa/Huggenes (5 to 10 mgC/L). Huggenes has a mixed agriculture/forest runoff which may explain that the ratio is slightly higher (ca 30%) compared to Støa (24%). The DOP to TDP ratio is considerably higher in the Dalen forest runoff compared to the agricultural sites (Søa, Huggenes). However, in absolute amounts, the concentration of DGT-DOP is considerably lower in the runoff from Dalen (1-5 µg P/L), compared to Huggenes with 4-12 µg P/L, Støa with5-20 µg P/L).
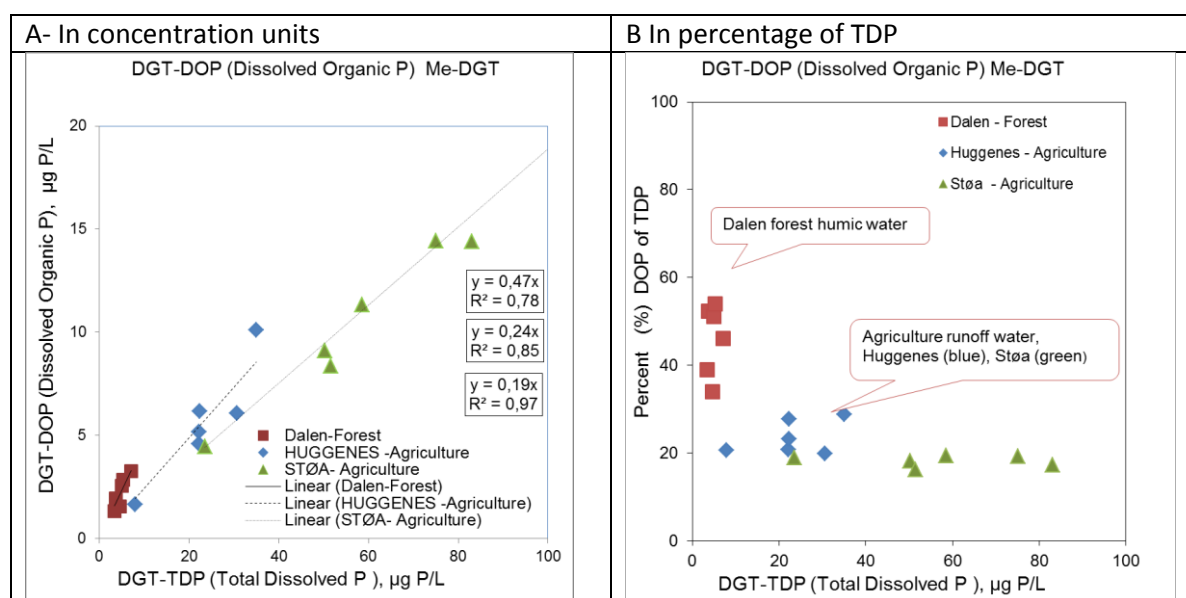
Figure 20

Distribution of DGT-DOP (Dissolved Organic P) relative to DGT-DTP (Dissolved Total P) in the three streams Dalen (forest runoff), Huggenes (agriculture/forest runoff) and Støa (agriculture runoff).

3.7.2 P:C ratios in the DOM material water from the Morsa catchments

Despite that dissolved organic matter (DOM) is the main natural transport mechanism of the micro nutrient phosphorous from terrestrial to aquatic environment, the knowledge regarding the content of P in the DOM is lacking. The Redfield atomic ratio1 06:16:1 of C:N:P, found in phytoplankton and throughout the deep ocean, are commonly also applied for aquatic DOM (Perdue, 2009, Spivakov et al. 1999). Current knowledge on the P content of DOM materials are summarized in Chapter 2.3. The C:P ratios found in DOM of natural water varies within broad ranges from less than the Redfield ratio of 106:1, up to as large as 15000:1. Typical number of are often an order of magnitude larger than the Redfield ratio, i.e. in the range 1000 to 2000.

Little information is available on the C:P ratio in dissolved organic material from the Morsa Vannsjø catchment, and in Norwegian DOM materials in general. To get a first impression the C:P

and P:C ratios were calculated based on the DOC in the water from the 3 stations above, and the DOP data from these DGT samples (Parekh 2012). The data are based only on the ca 20 DGT samples, so the results presented here are for information, and requires further investigations before clear conclusion can be drawn. The ranges for the C:P and P:C (%) ratios data are shown in Table 20 with explanation of how the calculation were performed, with a summary in Table 19. Expressed by the P:C ratios the DOM from the agricultural sites contains 5 to 10 times more P (0.02 to 1.4%) than the DOM from the forest site Dalen (0.001 -0.008%). The corresponding C:P ratios are in the range 10000 to 60000 from the forest site, and from ca 500 to 5000 from the agricultural sites.

This gives us 2 parameters for DOP fluxes in DOP budgets, i.e. the DOP/DTP ratios and the P:C (or C:P) ratios. The parameters obtained in Table 19 and 20 are first estimates based on this relatively limited dataset. These parameters can be improved by closer investigations of the whole EUTROPIA project database, which contains data for the TP, TDP, DIP, and DOP fractions in water (determined with the ordinary P fractionation methods). More modelling is possible based on these data, but this task is beyond the scope of this report which mainly deals with the use of DGTs. However, in this context it is noteworthy that the DGTs can display indication of these source patterns during the first study.

The phosphorus content of the DOM in water draining from forest soils is considerably lower than from the agricultural soils. The forested area constitutes 85% of the Morsa-Vansjø-catchment. Therefore, and as stated by Vogt (Vogt 2012, 2013), the total flux of phosphorus compounds from the forested areas is significant and must be taken into account when calculating the phosphorus budgets of the catchment. To get better figures for the parameters needed for such flux modelling, the data from the EUTROPIA project database mentioned above should be re-examined.

Table 19

C:P and P:C atomic ratios found in the stream water from the 3 stations based on data in Table 20.

Station	C:P atomic ratios	P:C atomic ratios in %
Støa (agriculture)	470 – 4850	0.02 – 1.4 %
Huggenes (agriculture/forest)	1000 -3700	0.03 - 0.10 %
Dalen (forest)	12000 - 59000	0.001 – 0.008 %

Table 20

C:P-atomic ratios and P:C atomic ratios (in%) in water runoff from the 3 streams. The calculations were performed as explained at the bottom of this Table, based on the DOC and the DOP fractions expressed in µEq/L of each to get atomic ratios.

Station		Input data	Input data		Output data		Output data	
		DOC	DGT-DOP		C:P atomic ratio Based on DOC/DOP		P:C atomic ratio Based on DOP/DOC	
		µEq C/L	µEq P/L	µEq P/L	C:P	C:P	P:C (%)	P:C (%)
			Low	High	Low	High	Low	High
Støa	25 percentile	423			2620	468	0,0382	0,2138
	Median	517	0.16	0,90	3203	572	0,0312	0,1748
	75 percentile	783			4852	866	0,0206	0,1154
Hug-genes	25 percentile	392			2428	1012	0,0412	0,0988
	Median	533	0,16	0,39	3302	1376	0,0303	0,0727
	75 percentile	601			3725	1552	0,0268	0,0644
Dalen	25 percentile	1594			41183	12355	0,0024	0,0081
	Median	1875	0,04	0,13	48438	14531	0,0021	0,0069
	75 percentile	2288			59115	17735	0,0017	0,0056

Explanation of how these data were calculated:

1. The input data for DOC (water) and DOP (by DGTs) are shown in the left column.
2. For the DOC concentrations, the medians, upper and lower quartiles (25 and 75 percentiles) at each site were used as input (Parekh 2012).
3. For the DOP fractions the lowest and highest values from the DGT-DOP fractions at each site in this work were used as input.
4. Each DOC value was divided by the lowest and highest DGT-DOP value to achieve a C:P atomic ratio or a P:C atomic ratio (%).
5. Thus a Low and High value for the C:P and P:C ratio could be achieved as shown in the output columns to the right.

3.8 Uncertainty of DGT measurements

The uncertainty of calculated DGT phosphorus fraction concentrations depends on those derived from the laboratory analytical procedures (DGT extracts) and those from the calculations using equation 3 in Table 7 An attempt is made in Chapter 4.7.2 to estimate the accumulated uncertainty through uncertainty budgets. A new uncertainty function is developed for the temperature correction of D during DGT deployments (Chapter 4.7.1).

3.8.1 Review of the temperature correction function for D

Equation 5 and 6 in Table7 are used to adjust D for temperature. The easiest way is to use the dimensionless D_t/D_o ratio to describe how D changes with temperature relative to the reference temperature (25^oC, where the ratio is equal to one, i.e. the diffusion coefficients for standard temperature can be applied). First the Stokes Einstein equation needs to be solved for D_t/D_o.

$$\frac{Dt}{Do} = \frac{T}{To} \times \frac{no}{n} \qquad \text{(Equation 5 in Table 7)}$$

The next step is to express the viscosity function of equation 6 for the dimensionless viscosity change ratio (compared to 25^oC) in the way required by equation 5 (*no/n*)

$$\frac{no}{n} = 10 \exp \frac{1{,}3702(t-25) + 0{,}00083(t-25)^2}{(109-t)} \qquad \text{(Equation 6 in Table 7)}$$

Combination of equation 5 and 6 express the ratio D_t/D_o. This ratio can now correct the diffusion coefficient to the required average sampling temperature.

$$\frac{Dt}{Do} = \frac{T}{To} \times 10 \exp \frac{1{,}3702(t-25) + 0{,}00083(t-25)^2}{(109-t)} \qquad \text{(Equation 5 and 6 combined)}$$

Table 20 shows the data for the D_t/D_o temperature correction factor from the combined equation 5 and 6. Figure 21A also shows the equation for the D_t/D_o factor versus temperature by a second order polynomial fit. No further improvements were achieved by third or fourth order polynomial fits. As shown in Table 20, the accuracy of a 2nd order curve is close to the derived from the original output from equation 5 and 6. Equation 10 expresses the new function for the D_t/D_o ratio versus temperature.

$$\frac{Dt}{Do} = 0{,}0001947 \cdot t^2 + 0{,}01716 \cdot t + 0{,}4492 \qquad \text{(Equation 10 in Table 7)}$$

The uncertainty of the D for a wrong average deployment temperature is given by the equation 11, derived from Figure 21B.

$$Uncertainty\ (\%) = 0.000419 \cdot t^2 + 0.054475 \cdot t + 3.765 \quad \text{(Equation 11 in Table7}$$

The uncertainty per degree (ºC) for each temperature unit varies from about 3.7 at 0 ^oC to 2.5 % at 30 ^oC. Assuming an uncertainty of 2 ^oC of the average DGT deployment temperature for a DGT deployment period, an average uncertainty per degree of 3 %, a total uncertainty of around 6 % is introduced.

A, Equation for the D_t/D_o correction factor

D_t/D_o 0 to 40 °C

$y = 0{,}0001946857x^2 + 0{,}0171622371x + 0{,}4492704439$

$R^2 = 0{,}9999999963$

♦ Dt/Do

—— Poly. (Dt/Do)

Temperature   °C

B Equation for the change in D_t/D_o factor in percent per °C,

D_t/D_o Percent change per °C from 0 to 40 °C

$y = 0{,}000419x^2 - 0{,}054475x + 3{,}765985$

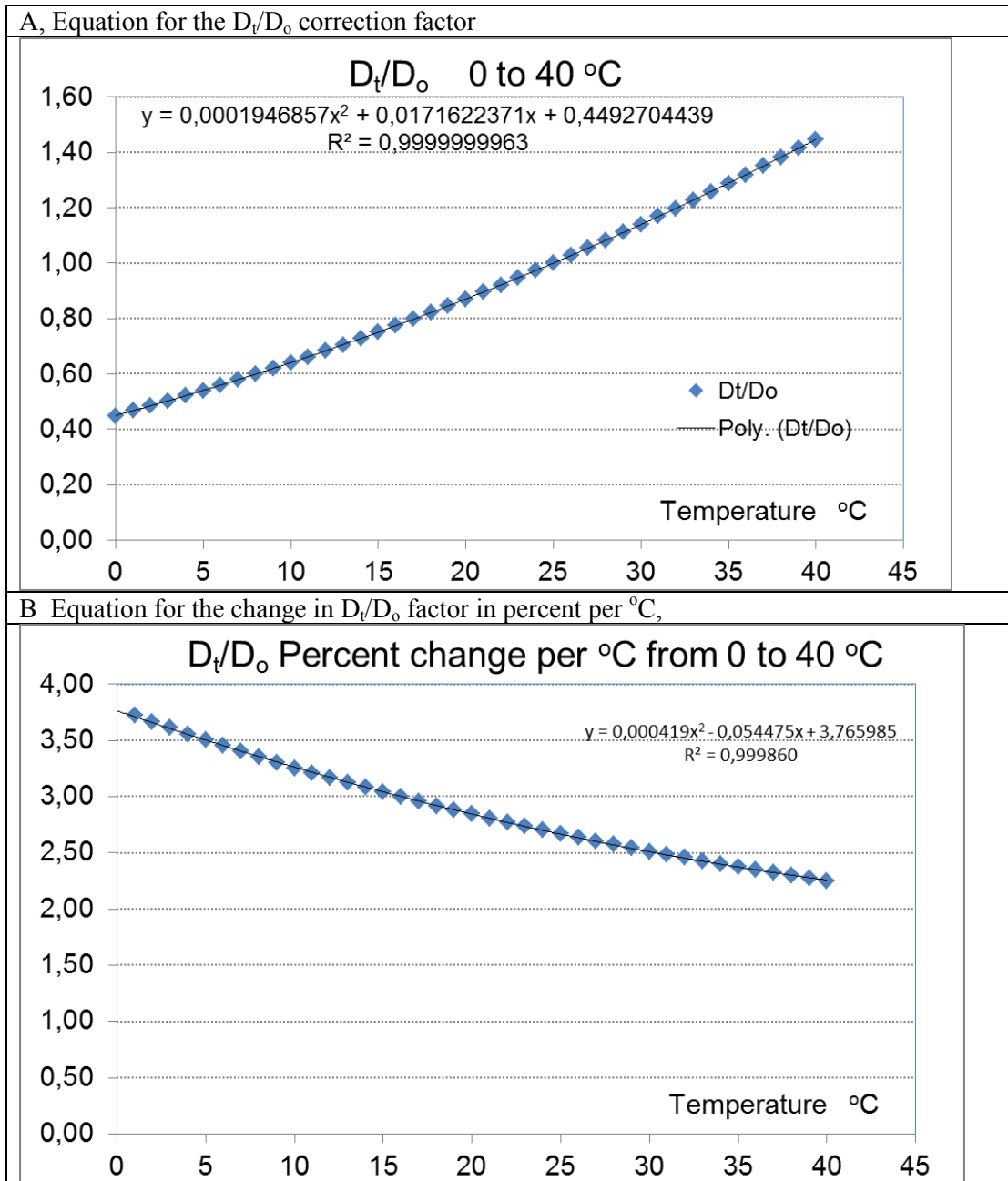$R^2 = 0{,}999860$

Temperature   °C

Figure 21.

D_t/D_o correction factor from 0 to 40 °C (A), and as change in the factor per °C in percent (B).(i.e. the uncertainty introduced due to a wrong estimate of the average deployment temperature).

Table 21.

Data for the function shown in Figure 21. Column 1 contains the D_t/D_o factor derived from the combined equation 5 and 6 in Table 7 Column 2 shows the result achieved with a 2nd order polynomial fit (Fig.20A). Column 3 shows the change of the D_t/D_o factor in step of 1° C relative to the preceding temperature,(ie $(D_{(t)} - D_{(t-1)})/D_o$) ,for example column(line30)-column(line29). Column 4 expresses the change $(D_{(t)} - D_{(t-1)})/D_o$ in percent (column 3) relative to the respective factor at the same temperature in column 1, for example 100*{column(line30)-column(line29)}/column (line29).

	D_t/D_o,	D_t/D_o	$(D_{(t)}/D_o) - (D_{(t-1)}/D_o)$	$\{(D_{(t)}/D_o) - (D_{(t-1)}/D_o)\}/D(t)$ in %¤
	Column 1	Column 2	Column 3	Column 4
Temperature	Derived from eq. 5 and 6, (Stokes Einstein + viscosity)	Output from 2nd order polynomial fit of Function in Figure 21A	Change per °C of D_t/D_o factor of column 1	Change per °C of factor in column 3 relative to column 1, expressed by Function in Figure 21B
oC	Unitless factor	Unitless factor	Unitless factor	Percent 100*(column3/column1)
40	1,4472	1,4471	0,0325	2,25
39	1,4147	1,4146	0,0321	2,27
38	1,3826	1,3824	0,0318	2,30
37	1,3508	1,3507	0,0314	2,32
36	1,3194	1,3193	0,0310	2,35
35	1,2885	1,2883	0,0306	2,37
34	1,2579	1,2577	0,0302	2,40
33	1,2277	1,2275	0,0298	2,43
32	1,1978	1,1977	0,0294	2,46
31	1,1684	1,1683	0,0290	2,49
30	1,1394	1,1392	0,0287	2,51
29	1,1107	1,1106	0,0283	2,54
28	1,0825	1,0823	0,0279	2,58
27	1,0546	1,0545	0,0275	2,61
26	1,0271	1,0270	0,0271	2,64
25	1,0000	0,9999	0,0267	2,67
24	0,9733	0,9732	0,0263	2,70
23	0,9470	0,9469	0,0259	2,74
22	0,9211	0,9210	0,0255	2,77
21	0,8955	0,8954	0,0251	2,81
20	0,8704	0,8703	0,0248	2,84
19	0,8456	0,8455	0,0244	2,88
18	0,8212	0,8212	0,0240	2,92
17	0,7973	0,7972	0,0236	2,96
16	0,7737	0,7736	0,0232	3,00
15	0,7505	0,7504	0,0228	3,04
14	0,7277	0,7276	0,0224	3,08
13	0,7053	0,7052	0,0220	3,12
12	0,6833	0,6832	0,0216	3,17
11	0,6616	0,6615	0,0212	3,21
10	0,6404	0,6403	0,0209	3,26
9	0,6195	0,6194	0,0205	3,30
8	0,5990	0,5989	0,0201	3,35
7	0,5790	0,5789	0,0197	3,40
6	0,5593	0,5592	0,0193	3,45
5	0,5400	0,5399	0,0189	3,50
4	0,5211	0,5210	0,0185	3,56
3	0,5025	0,5024	0,0181	3,61
2	0,4844	0,4843	0,0178	3,67
1	0,4666	0,4666	0,0174	3,72
0	0,4492	0,4492		

3.8.2 Uncertainty budget for the DGT sampling process

The total uncertainty of a measurement process can be estimated by the error accumulation law. This is achieved by summing up all the squared standard deviations of each independent input component contributing to the uncertainty of the sampling, analytical and calculation process. It is common to change all uncertainty components to a dimensionless value, such as by the coefficient of variation (CV%) which is the ratio of the standard deviation of each uncertainty component divided by the mean value (Konieznik and Namiesnik, 2009). With all standard deviations in the same unit the modelling can now be done in a spreadsheet. All CV% are squared, the Sum of Squares (SS) of all constituents is calculated, and the total standard deviation in percent is derived as the square root of the SS of all components.

With all the input parameters in the spreadsheet model it is possible to estimate the total uncertainty. Furthermore it is possible to identify the factors having the largest influence on the total standard deviation, and thereby the total uncertainty. The uncertainty budgets are divided into at least three sub-processes which are included in the uncertainty calculations shown in Table 22, 23 and 24.
1. Laboratory procedures related to the measurement in the laboratory
2. Calculation of the DGT concentrations based on all the separate calculations processes in the DGT equation.
3. The field sampling process. The uncertainty of the sampling process is related to the collection of a representative sample. This uncertainty component may vary within broad ranges depending of the variability of the concentrations at the site, and needs separate assessments. This topic is therefore not discussed here, as the contribution depends on the sampling conditions.

To estimate the total uncertainty a coverage factor is usually added on top of the total standard deviations (Konieznik and Namiesnik, 2009). A coverage factor is not used in this evaluation. Instead two estimates are chosen, one Low and one High, to get an impression of the range of expected uncertainties. The most important purpose of an uncertainty budget is to identify the factors having the largest influence of the total uncertainty. By this budget it is possible to model how the total uncertainty changes by method improvements. The natural approach is to focus on the factors having the largest contributions to the total uncertainty. By this also cost benefit analysis can be included to evaluate the cost of the efforts compared to the benefit of the improved accuracy.

This budget process is divided into 3 parts. In Table 22 all the uncertainty components are evaluated and given an estimate for a Low and High CV value. In Table 23 the different components are squared and summed. The components are divided into 3 subgroups. Those derived from the laboratory processes and the general DGT calculation processes which are the same for both the DIP and DOP fractions. The uncertainty for the components connected to the Diffusion coefficients are different for the DIP and DIP fractions. These are evaluated separately. The SS of each subgroup is collected in Table 24. The total uncertainty for the DIP and DOP fractions are calculated as the square root of the SS. By this the total uncertainty if achieved for the DIP and DOP fractions.

Table 22
Evaluation of the uncertainty components of the laboratory processes and the DGT calculation processes. All the evaluations for the DGT components are for the APA membrane.

Laboratory processes	
Gravimetry	Most gravimetric processes are very well controlled by modern high precision balances. Low/High estimates of 0.1 and 0.5 % are chosen.
Volumetry	Volumetric processes are also well controlled by equipment with uncertainty specification below 0.5%. However, modern micropipettes are often used. These have higher uncertainty. Low/High estimates of 0.5 and 1.0 % are chosen.
Analysis	Based on experience with quality control samples of the CFA analyser, Low/High estimates of 2 and 5% are chosen.
Other	This relates to processes in the laboratory not quantified. Low/High estimates of 1 and 3 % are chosen.
DGT calculation processes	
Temperature	The deployment temperature is calculated either as the average temperature in ^{o}C determined at start and stop, or as the average temperature obtained by temperature loggers. By using a temperature logger the uncertainty of the average temperature can be kept below 1 ^{o}C. This uncertainty is expressed by equation 11 in Table 6, which predicts an uncertainty between 3.5 and 2.5 % per ^{o}C in the range 0 to 25 ^{o}C. For simplicity an uncertainty of 3 % per degree C is used. This uncertainty component depends on the temperature fluctuations at the site. In large water bodies (lakes, coastzones, the open ocean) the temperature fluctuations are often within a few degree C for deployment periods of 1 to 2 weeks. In smaller streams day-night fluctuations can be in the order of 5 degrees and lead to larger uncertainties, based on only start –stop measurements. However, assuming an uncertainty of the average temperature is in the range 1 to 2 ^{o}C, this corresponds to a Low/High estimate of 3 and 6 % which is chosen in Table 23. This is the same for both the DIP and DOP fractions.
The diffusion length (L).	The default total length of L of the diffusion path of the DGT sampler is 0.11 cm (Table 5). The uncertainty of L is mainly related to the DBL term. This is treated as a separate uncertainty component below. However, the membrane thickness has also an uncertainty component of 0.02 to 0.05 cm from the production and gel swelling process. Compared to the default L of 0.11 cm, a Low/High estimate of 2 and 5 % is chosen for this uncertainty component. This is the same for both the DIP and DOP fractions.
The diffusive boundary layer (DBL).	Garmo, Naqvi et al. (2006) revised the DBL model, and Warnken, Zhang and Davison (2006) investigated the uncertainties related to the DBL. Figure 22 show the relation between DBL and water flow velocity in cm/sec, predicting at DBL of ca 0.1 mm at 10 cm/sec. From 10 to 2 cm/sec the DBL increases from 0.1 to 0.2 (or 0.3 mm depending on the DBL model used). Below 1 cm/sec the increase is very steep. Above 2 cm/sec the uncertainty of the DBL estimate is in the order or 0.1 mm. Several estimates of the DBL layer have been published, but these have not been related to the water flow velocity vs DBL function as shown in Figure 22. The DBL measurements have been conducted by the reciprocal mass plot model (equation 8 in Table 6) using DGT deployments with two or three membrane thicknesses (0.8 to 2 mm) without determining the water flow rates. In the recent study of antibiotics in wastewater by Jones, Chen et al. (2013) a DBL of 0.23 mm was reported by the reciprocal plot method, but without reporting the water flow velocity or an uncertainty estimate. A moderate water flow velocity between 2 and 10 cm/sec is chosen the DBL is ca 0.01 cm with an uncertainty 0.05 to 0.1 mm. Compared to the L term of 0,11 cm, a Low/High estimate of 5 and 10 % is chosen for this uncertainty component. This

	uncertainty is the same for both the DIP and DOP fractions.
Area of the sampler window (A)	The window diameter is 20 mm with a window area (A) of 3.14 cm^2. The uncertainty of the diameter has not been specified. If the uncertainty is estimated to 0.2 mm, the area will vary from 3.08 to 3,20 cm^2, i.e. an uncertainty of ca 0.05 cm^2 (1.5%). A Low/High estimate of 1 to 3 % is chosen. This uncertainty is the same for both the DIP and DOP fractions.
Effective Area of the sampler window (A)	Around the rim of the sampler window a lateral diffusion process is occurring both in the DBL layer in front of the window and in the membrane behind, as described and modelled by Warnken et al (2006). The net effect is that the apparent window becomes larger. According to Garmo (2014) the window will be ca 3.8 cm^2 compared to the nominal area of 3.14 cm^2, i.e. a difference of ca 0.07 cm. The lateral process in the membrane behind the window is counteracted by the DBL layer in front of the window, which reduces this area effect, and the corresponding uncertainty. An estimate between 0.15 to of 0.30 cm^2 is chosen for this process which gives a Low/High uncertainty estimate of 5 to 10 %. This uncertainty is the same for both the DIP and DOP fractions.
The diffusion coefficient (D). DIP	For the orthophosphate species the estimate for D is assumed to have a good accuracy, according to the good agreement by the D values obtained by Zhang and Davison (1998) and Røyset, Sogn et al (2004). When the pH correction function of this work is used, the accuracy is further improved. For this uncertainty calculation Low/High estimates of 3 and 5 % is chosen.
The diffusion coefficient (D). DOP	For the DOP fraction the uncertainty depends on the molecular weight distributions as discussed in chapter 4.3. The uncertainties will be from 10 to 50 % depending on how detailed information of the Mw distribution is available. Only rough estimates can be given and a Low/High estimate of 10 % and 30 % is chosen.
Membrane resistance term (R)	The uncertainty of R is expected to be low for the ortophosphate species of the DIP fraction. A Low/High value of 3 and 5 % is chosen.
Membrane resistance term (R)	The uncertainty of R for the DOP fraction depends on the molecular weight distribution as shown in Table 15.The knowledge is limited for the time being. Only rough estimates can be given and a Low/High value of 5 and 10 % is chosen.
pH	The pH dependence of D is covered by the new equations in chapter 4.2. For the DIP fraction the D changes with ca 0.15 per pH unit, corresponding to an uncertainty of ca 3% introduced per pH unit. For the DOP fraction the D has an uncertainty of ca 0.1 per pH unit in the range 3 to 7 (chapter 4.3). To cover this uncertainty component a Low/High estimate of 1 and 3 % is chosen.
Biofouling	During long deployment times, algal and bacterial growth on the membrane surface may produce biofilms which increase the total diffusion path (the L term) Biofouling occur usually at deployment times above 1 to 2 weeks. For the time being we do not have good numbers for the uncertainty due to the biofilm thickness, and this is not taken into account in this uncertainty evaluation.

3.8.3 Evaluation of the uncertainty budgets

The laboratory processes
The uncertainty contributions from the gravimetric and volumetric operations are generally below ca 1 %. The largest contribution comes from the analysis by the CFA analyser. According to these estimates the contributions from the laboratory processes lie in the range from 2 to 6%, which is low compared to the total uncertainty budgets from ca 10 to 38 %

The DGT calculations without the diffusion coefficient
The uncertainty contribution is low from the elution efficiency, the temperature, the membrane thickness and the window area have. If these are summed up separately, they contribute to ca 4 to 9 % ((CV)^2 of 15 and 79).

The largest influence comes from the DBL and the effective window area. These factors contribute between 7 and 14% .((CV)^2 of 50 and 200). The DBL contributes 5-10% alone. The effective window area depends on the thickness of the boundary layer as the diffusion in front of the filter When the DBL is reduced, i.e. at higher water flow velocities, the diffusion length in front of the filter decreases, and the influence of the effective window area is reduced.

The DGT calculations related to the diffusion coefficient
For the DIP fraction the contribution from the D, R and pH is relatively low in the range 4 to 8%.

For the DOP fraction the uncertainty contribution from the D, R and pH has the largest influence with estimates between 11 and 32%. These 3 factors are the dominating uncertainty components of the total uncertainty budget of 14 to 37% for the DOP fraction.

Clearly, this is the largest uncertainty for the accuracy of the DGT-DOP fraction. To improve the accuracy we need more accurate and appropriate values of D and R. More information about the Mw distributions is needed to achieve the most appropriate D and R values (based on either an average Mw, or as a Mw distribution curve). If a Mw distribution curve is determined, the Mw specific D and R values of Table 15 can be used.

Total uncertainty
For the DGT-DIP fraction the total uncertainty estimates are between 9 to 19%.
For the DGT-DOP fraction the total uncertainty estimates are from 14 to 37%.

Sampling precision obtained in the field campaign
In chapter 4.5 (Table 17) a total sampling precision of ca 13 % RSD (~4 µg P/L at ~30 µg P/L) was achieved for the DGT-DIP fraction. This is within the total uncertainty of 9 to 19 for the DIP fraction above.

A RSD of 18-20% (~2 µg P/L at ~10 µg P/L) was achieved for the DGT-DOP fraction. This is within the estimates of 17 to 37% from Table 24. A high sampling precision is possible to obtain, if the standard deviation of 2 µg P/L is representative for the fairly low concentration range 1 to ca 30 µg P/L found for the DOP fraction at these sites.

Uncertainties of the DGT samplers compared to grab sampling
Within the EUTROPIA project relatively large deviations (by a factor of 1 to 2, i.e. 100 to 200%) were observed between the results reported for water grab samples analysed at different laboratories for the phosphorus fractions TP, TDP and DIP (Parekh, 2012). Large deviations were also found in a recent laboratory comparison of water samples added suspensions of soil and sediment materials for the parameters suspended solids (SS), TP and DIP (Krogstad, Øygaard, & Skarbøvik, 2013). In relation to this, the estimates of 10 - 30% uncertainty for the DGT samplers are not bad. The information value of a measurement with a standard deviation of 10-30 %

(between 2 and 5 µg P/L, as achieved in Chapter 4.3), is thus still very useful. Moreover, the DGT produce new data for the DOP fractions which has not previously been available.

Assessments of possibilities for improvement
The uncertainty of the DBL component can be improved by ensuring that the water flow velocity is kept above at least 1 cm/sec, and preferably above 5cm/sec. Above 5 the uncertainty of the DBL is reduced to below 0.01 cm (0.1 mm). The uncertainty may be further reduced at higher water flow velocities, which also reduce the uncertainty of the effective window area component. Thus the uncertainty from these two components could be reduced to at least 10%.

Fortunately water flow velocities in streams and rivers are generally above 10 cm/sec. At lake surfaces the wave motion give satisfactory water movement in most cases. A higher DBL-layer uncertainty may be expected at low water perturbation, such as below the wave motion area in lakes and ponds. In such situations satisfactory DGT sampler movement may be achieved by connecting the DGT samplers to surface buoys which is kept in motion by the wave actions.

The achilles heel of the DGT-DOP sampler is the needs for more accurate diffusion coefficients for the molecules in the DOP fraction. Better information about the molecular weight distribution of the DOP fraction is needed to be able to estimate Mw specific D and R values, and a revised and improved Table 15 may be developed (see also chapter 5.2).
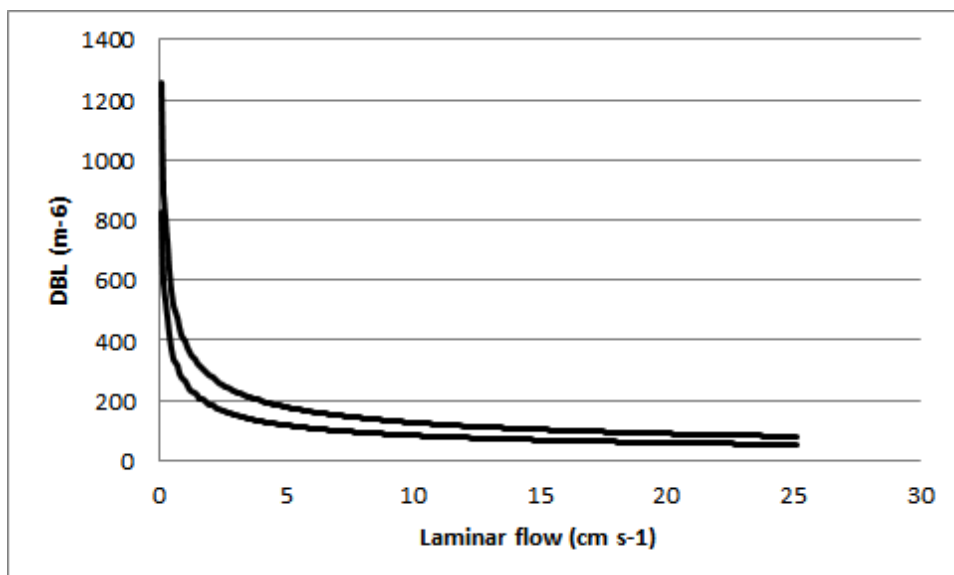

Figure 22.
Visualization of the Diffuse Boundary Layer (DBL in µm) vs. water flow velocity based on the model by Garmo, Naqvi et al. (2006) (lower curve) compared to the original one (upper) by Davison and Zhang (1994).

Table 23.
Uncertainty budget for the DGT sampler based on the different uncertainty components of the analytical and DGT calculation process, expressed as CV (coefficient of variance %). The input values for the uncertainty are divided into a Low and High estimate for the CV%. For the diffusion coefficient separate calculations are done for the DIP and DOP fractions.

		Coefficient of variation (CV)		Squared (CV)^2	
		Low	High	Low	High
Laboratory procedures					
DIP/DOP	Gravimetry	0,1	0,5	0,01	0,25
DIP/DOP	Volumetry	0,5	1	0,25	1
DIP/DOP	Analysis by CFA	2	5	4	25
DIP/DOP	Other analytical	1	3	1	9
DIP/DOP	**Sum of squares (SS)**			**5**	**35**
DGT calculation without the Diffusion coefficient					
DIP/DOP	Elution efficiency	1	3	1	9
DIP/DOP	Temperature	3	6	9	36
DIP/DOP	Window Area	1	3	1	9
DIP/DOP	Membrane plus filter thickness	2	5	4	25
DIP/DOP	Effective window area	5	10	25	100
DIP/DOP	Diffusive Boundary Layer	5	10	25	100
DIP/DOP	**Sum of squares (SS)**			**65**	**269**
Diffusion coefficient of the DIP fraction					
DIP	Diffusion coefficient	3	5	9	25
DIP	Membrane resistance	3	5	9	25
DIP	pH dependance of D	1	3	1	9
DIP	**Sum of squares (SS)**			**19**	**59**
Diffusion coefficient of the DOP fraction					
DOP	Diffusion coefficient	10	30	100	900
DOP	Membrane resistance	5	10	25	100
DOP	pH dependance of D	1	3	1	9
DOP	**Sum of squares (SS)**			**126**	**1009**

Table 24.
Summary of the uncertainty budget for the DGT sampler based on the different uncertainty components of the analytical and DGT calculation process in Table 23. The input values for the uncertainty are divided into a Low and High estimate for the CV as explained in the text.

Fraction	Process	Sum of Squares (CV)2		Uncertainty (CV)	
		Low	High	Low	High
DIP/DOP	Laboratory processes	5	35	2	6
DIP/DOP	DGT calculations without D	65	269	8	17
DIP	Diffusion coefficient DIP	19	59	4	8
DOP	Diffusion coefficient DOP	126	1009	11	32
DIP	Sum uncertainty of the DIP fraction	89	373	9	19
DOP	Sum uncertainty of DOP the fraction	196	1348	14	37

4. CONCLUSIONS

4.1 Analytical performance

This study of DGTs for collection of dissolved inorganic and organic phosphorus fractions (DIP and DOP) in water has many promising results. The most important analytical performance properties of the DGT sampler are summed up in Table 25, and discussed below.

Table 25.
Overview of important performance parameters of the Me-DGT and Fe-DGTs samplers used for sampling of the TDP, DIP and DOP fractions in freshwater.

Parameter, fraction	Sampler	Performance figures
Extraction efficiency	Fe-DGT	100 % for TDP and DIP fractions (one extraction needed).
Extraction efficiency	Me-DGT	87 ($\pm$ 4) % for TDP and DIP fractions. Two extractions are recommended.
Adsorption efficiency	Fe-DGT Me-DGT	100 % for Mw fractions of DOP up to 1000 Da. Above this Mw less information is available
Sampling precision	Fe-DGT Me DGT	$\sim$ 13 % for DGT-DIP $\sim$20 % for DGT-DOP
Adsorbent capacity	Fe-DGT	Ca 7 μg as P (Davison and Zhang, 1998) 15-25 μg as P (Panther et al. 2011, 2013)
Adsorbent capacity	Me-DGT	Ca 37 μg as P (Panther et al., 2011)
D for DIP fraction	Fe- DGT Me-DGT	5.5 (10^{-6} cm^2/sec) at pH 7 for the ortho-phosphate species.
pH correction of D for DIP fraction	Fe-DGT Me-DGT	According to Figure 14, and Equation 13 in Table 7 (Ortho-phosphate species), D = - 0,15pH + 6.62 at R=30%
D for AMP	Fe-DGT Me-DGT	3.2 ($\pm$ 0.5) 10^{-6} cm^2/sec 4.0 10^{-6} cm^2/sec (Buffle et al. (2007)).
D for IP6	Fe-DGT	1.4 ($\pm$ 0.2) 10^{-6} cm^2/sec 3.0 10^{-6} cm^2/sec (Buffle et al. (2007)).
R (for DIP fraction	Fe-DGT Me-DGT	Membrane resistance estimated to 30 % (Davison and Zhang, 1998). Agrees with results in this work
D for the DOP fraction	Fe-DGT Me-DGT	3.0 to 4.0 10^{-6} cm^2/sec as first approximation. Depends on molecular weight and membrane resistance. More validation work is needed which is combined with the same for the R factor below.
R for the DOP fraction	Fe-DGT Me-DGT	A relation between the membrane resistance R and Mw is obtained according to Table 15. More validation work is needed.

DGT allow the collection and subsequent determination of 2 phosphorus fractions, DGT-DIP and DGT-DOP, which can be handled separately with separate diffusion coefficients (D). Both the Fe and Me-DGTs collected both fractions. After extraction it was possible to separate the DIP and DOP fractions from each other by the CFA auto-analyser method developed at NIVA.

Low Molecular Weight (LMW) Organic phosphorus molecules (AMP and IP6) with Mw of347 and 736 Da showed linear uptake indicating quantitatively adsorption by both the Fe- and Me-DGTs adsorbents. The diffusion coefficients of 3.2 and 1.3 (10^{-6} cm^2/sec) were 20 % and 60 % lower than those calculated by Buffle (4.0 and 3.2 respectively). This difference is due to membrane resistance in the DGT membrane of the charged molecules. The linear uptake indicate quantitative adsorption of these LMW model compounds.

The extraction efficiency by sulphuric is 100 % for the Fe-DGT adsorbent AMP, IP6 and DOP compounds collected in the field. For the Me-DGT adsorbent the extraction efficiency by 1 M NaOH is 87% both for IP6 in the laboratory and for DOP compound sampled in the field. Two repeated extractions are recommended for the Me-DGT.

The total adsorption capacities are in the range 7 (Fe-DGT) to 37 µg P (Me-DGT) of the adsorbents. The adsorbent can be loaded to 50% capacity before saturation/overloading occurs (Davison and Zhang 1998). At a DIP concentration of 10 µg P/L and weekly sampling (sampling volume ca 0.1 L), the amount sampled is 1µg P. This is ca 10% of total capacity for a Fe- and ca 3% of a Me-DGT. 2 week deployment periods are acceptable, but at TDP concentrations above 100 µg P/L the deployment times should be kept below 7 days to prevent risk for overloading.

Earlier the Metsorb™ adsorbent was considered to have higher capacity for ortho-phosphate than ferrihydrite. Lately Price, Teasdale et al. (2013) questioned this. The capacity may be increased by using larger amounts of ferrihydrite in the adsorbent layer. This may require improvements in the casting technology but is worth the efforts as ferrihydrite is a much more convenient adsorbent as it dissolves in dilute sulphuric acid, giving an ideal matrix for the simultaneous determination of the TDP, DIP and DOP- fractions by CFA analysis. This extract is also useful for further separation and analysis of the collected DOP molecules (Mw, charge, C:P ratios etc).

Both the Fe- and Me-DGT samplers showed good performance in a field study of DIP and DOP fractions in stream water in the Morsa-Vansjø catchment. The sampling precision was best for the DGT-DIP fraction (~13 % RSD) while ~20 % for the DGT-DOP fraction.

AMP and IP6 are stable on both the Fe-DGT and Me-DGT adsorbents during storage times of several weeks. The same good stability is expected for DOP compounds from field samples (as demonstrated by storage of exposed DGT for up to 8 weeks). A test of maximum storage time is still lacking, but 4 week at $4^{o}C$ is assumed to be safe. The same is the case in the acid preserved extract from both DGT adsorbent. Morleghem et al. (2013) also confirmed this good stability in a study of a number of low molecular weight organic phosphorus compounds in acid media at pH around 1.

The uncertainty budgets developed in chapter 4.7 shows a total uncertainty from 10 to 40%.

The largest analytical challenge for the DGT sampler lies in the estimate of the diffusion coefficient (D). For the ortho-phosphate species in the DGT-DIP fraction a new correction function was developed where the numerical value of the D value decreases with 0.15 per pH unit. The earlier the diffusion coefficient for the $H_2PO_4^-$ -specie has been reported to 6.07 (10^{-6} cm^2/sec). For field measurements a D value of 5.5 at pH of 7.2, gave the best fit to the data, indicating that the pH correction improves the accuracy of the DIP fraction. The uncertainty of the D could be reduced to below 5% by this correction.

For field sampling a D value for the DGT-DOP fraction was initially considered to be 3.0 (10^{-6} cm^2/sec). The uncertainty of this estimate is relatively large, and may be in the range of 0.5 to 1.0 (10^{-6} cm^2/sec), depending on the molecular size distribution of the compounds in the DOP fraction The modelling in chapter 4.5, showed that the best fit was achieved at a D value of 3.7 (10^{-6} cm^2/sec) in the agricultural runoff water. This indicates that the DOP compounds in the agricultural runoff waters collected by the DGT may belong to a fairly low molecular weight fraction with Mw predominantly between 300 up to 1000 Da. However more research is needed in this are to clarify these preliminary assumptions.

The membrane resistance (R) vs log(Mw) curve in Figure 12 may be used to find the R factor to correct the D value for the DOP fraction. The challenge is to achieve data for the molecular weight

distribution of the DOP compounds in the water samples. With this information, the uncertainty of the average diffusion coefficients may be reduced, for example by dividing the DOP fraction into molecular weight fractions (as proposed in Table 15).

The DGT is a very promising tool for the sampling of dissolved inorganic phosphorus (DIP) fractions in water during environmental monitoring in general, in eutrophication studies in special, and for developments of environmental technologies. The improved accuracy of the D of the orthophosphate species makes the DGT-DIP fraction almost as accurate as normal water DIP methods. A major benefit is the in-situ separation of dissolved and colloidal/particulate fraction by the 5 to 10 nm pore size of the diffusion membrane. Another useful feature is the average concentrations achieved, which may be more relevant than grab sample values for establishing threshold values of ortho-phosphate species to predict algal blooming.

4.2 Future research

The DGT sampler appears to have potential as a new *in-situ* sampling and separation tool of the DOP fractions. The preliminary promising results and experience gained in this work is important and points out several future research directions:

4.2.1 Molecular weight distribution of the DOP fraction

Only one D value can-not cover the whole molecule weight range of the DOP fraction from 300 to 10000 Da. The best way forward is to separate the DOP fraction into 3 to 5 molecular weight ranges such as LMW, MMW, and HMW fraction as proposed in Table 15. . By this the different molecular weight fractions can be assigned individual D and R values.. This requires molecular weight separation methods such as size exclusion chromatography (SEC), ultrafiltration (UF), Field Flow Fractionation (FFF) or conventional HPLC techniques etc. These techniques may also be connected to ICPMS or MS, so that the amount of phosphorus in each fraction may be determined. This may appear to be a time consuming approach. However, since the molecular weight distribution of the DOP fraction may be fairly stable over time at the same site, only a few molecular weight distribution spectra may be sufficient to characterize the DOP fraction at each site. Thus much more accurate data for the DOP fraction may be achieved, than the total values for the DOP fraction as used in this work.

4.2.2 Adsorbent collection efficiency and capacity.

The molecules of the DOP fraction are expected to have similar properties as common NOM/DOM molecules. The phosphate groups may not be so exposed to the outside of the molecule, so that the adsorption is directly caused by the phosphate group. The DOP molecules are predominantly negatively charged, and may also be adsorbed on these adsorbents simply by negative charges. The adsorption of the DOP fraction may rely on both phosphate and anion adsorption mechanisms. These are not fully understood and require more research. Moreover, the adsorption capacity of the ferrihydrite adsorbent used at present is lower than desired. Teasdale et al (2013) showed that the adsorption capacity of this adsorbent could be increased by simple measures. A high capacity ferrihydrite adsorbent should be produced.

4.2.3 Can membrane resistance (R) be predicted based on Mw?

The membrane resistance (R) curve in Figure 12 shows an almost linear relationship between R and log(Mw), both for the ordinary APA (agarose polyacrylamide) and the AGE (agarose) based DGT membranes. Table 15 summarizes the R values for separate Mw ranges from 300 to 10000Da. These data are based on only a few humic and LMW organic compounds. To get more accurate data, the R for a few more LMW (300 to 1000 Da) and HMW compounds (1000 -10000

Da) should be tested. Such data are rather easily to achieve by diffusion cell test of the respective membranes. Preferably the DGT community should agree on a set of ca 10 test compounds from 300 to 10000 Da for testing of new membrane. The need for R values will increase when the DGT technology now "goes organic" with sampling of organic compounds such as antibiotics, pharmaceuticals, personal care product etc.

4.2.4 pH effects

The D of the orthophosphate species change with a numerical value of -0.15 (R=30%) per pH unit (Figure 14). Similar pH effects may also occur in other oxyanions with hydrolysis such as As(V) and Se(VI). The diffusion coefficients of fulvic and humic acids decrease with a numerical value of ca 0.5 units from pH ca 7 to 3 due to agglomeration to larger molecular entities when pH decreases (Lead et al. (2003), Buffle et al (2007). Other LMW organic compounds such as antibiotics, pharmaceuticals, personal care products (PCP) ect, are mainly polyprotic molecules where the charge increase with pH. The pH dependence of D for organic molecules need further research to examine how the D changes with pH. This is of relevance for the organic compounds above as well as for the organic molecules of the DIP fraction.

4.2.5 New information needed when the DGT moves to collection of organic compounds

This new knowledge is particularly useful for further developments of the DGT for the sampling of the DOP fraction in water. This is also very relevant for the DGT community where the DGT research now is moving towards the sampling of organic compounds in water. Chen et al. (2012), Jones et al.(2014) started this research area on the collection of antibiotics, personal care products and pharmaceuticals in wastewater. Very promising results have been achieved. This research need the same knowledge development of how the D values can be estimated and corrected for by the properties of the membrane the resistance, pH dependence, and other molecular properties as discussed in this work.

4.2.6 Can the DGT collect the most bioavailable fraction?

The APA membrane collect the LMW and medium molecular weight (MMW) fraction <5 kDa with higher efficiency than the HMW fraction above 5 to 10 kDa. This LMW and MMW group of the DOP fraction is also expected to be the most **bioavailable organic molecules** as nutrient source by microorganisms and for enzymatic degradation as pointed out by Worsfold et al (2008). If the DGT collect this bio-available MW fraction of the DOP compound with higher efficiency than the HMW fraction, we may have got grip of a simple and powerful *in-situ* separation tool with large potential for future research. DGTs with this cut-off (or with membranes with customized cut-off) can be used to monitor mineralisation rates of DOP compounds to ortho-phosphate species in manures, sludges, soils etc. by new environmental technology methods sludges (such as proposed by Eggen, 2013).

4.2.7 Customized pore-sizes of membranes

The pore size and membrane resistances of different membrane types, makes it possible to develop DGT samplers with different in situ separation properties. Open pore agarose membranes has low membrane resistance collect while the most constricted polyacrylamide based membranes excludes almost all molecules above 1000 Da (such as the CGA membrane in Table 14). Many separations may thus be achieved *in-situ* by using 2 to 4 membrane types.

4.2.8 More information about the DOP, DIP and TDP fractions needed for P budgets

Some interesting source patterns of the TDP, DIP and DOP fractions and C:P ratios were observed in stream water from the forest and agricultural areas in the Morsa Vansjø catchments (as discussed

in chapter 4.6). The DOP/TDP ratio in stream water draining an agricultural field was 20 to 30%, while this ratio was 40 to 60 % in the water from an acid forest soil site. The C:P ratios were lower in the water from the agricultural soil than those from the forest soils. The concentrations of the DIP and DOP phosphorus fractions were considerably lower in the runoff from the forest soils than from the agricultural soils. Nevertheless, since the forests cover 85% of the Morsa-Vansjø catchment, this flux or organic phosphorus compounds may be considerable. Closer evaluation of the budgets is necessary in order to quantify the fluxes of the different phosphorus fractions from the different landscape types, and especially the DOP fractions. Additional data to support such calculations are available in for example the EUTROPIA project, which now deserves further investigations.

5. ACKNOWLEDGEMENTS

6. References

Beckett, R; Jue, Z; Giddings, J.C (1987), Distributions of Fulvic and Humic Acids Using Flow Field-Flow Fractionation., Environ. Sci. Technol., 21, 289-295.

Blankenberg, A.; Turtumøygard, S.; Pengerud, A.; Borch, H.; Skarbøvik, E.; Øygarden, L.; Bechmann, M.; Syversen, N.; Vagstad, N. Tiltaksanalyse for Morsa: "Effekter av fosforreduserende tiltak i Morsa 2000-2006"; Bioforsk Jord og Miljø Ås, Norway, 2008; p 54.

Buffle (2014), pers. comm.

Buffle, J.; Zhang, Z.; Startchev, K.(2007), Metal Flux and Dynamic Speciation at (Bio) interfaces. Part I: Critical Evaluation and Compilation of Physicochemical Parameters for Complexes with Simple Ligands and Fulvi/Humic Substances. Environmental Science & Technology 2007, 41, (22), 7609 - 7620.

Buffle J, (1988) Complexation Reactions in Aquatic Systems: An Analytical Approach, Ellis Horwood series in analytical chemistry, Chichester, Uk, 361 pp.

Jones K.C., Chen, C., Zhang, H. (2012) A novel passive water sampler for in situ sampling of antibiotics, J. Environ. Monit. 2012, 14, 1523 -1530.

Chen, C., Zhang, H., Ying, G., Jones, K.C. (2013) Evidence and recommendations to support the use of a novel passive water sampler to quantify antibiotics in wastewaters, Environmental Science and Technology. 47, 23, p. 13587-13593.

Cleveland, C. C. & Liptzin, D. 2007. C:N:P stoichiometry in soil: is there a ''Redfield ratio'' for the microbial biomass? Biogeochemistry, 85, 235–252.
Cussler, E. L., Diffusion: mass transfer in fluids systems. 3 ed.; Cambridge University Press: 2009.

Costello, A.J.R., Glonek, T. & Myers, T.C. (1976) 31P nuclear magnetic resonance-pH titrations of myo-inositol hexaphosphate. Carbohydrate Research 46: 159-171.

Cussler, E. L., Diffusion: mass transfer in fluids systems. 3 ed.; Cambridge University Press: 2009.

Dahl, I (2012) Sammenlignende laboratorieprøvninger (SLP) – Analyse av ferskvann. SLP 12-21. NIVA report 6462-2012, Norwegian Institute for Water Research, NIVA , Oslo.

Davison, W.; Zhang, H. 1994, In-situ speciation measurements of trace components in natural waters using thin-film gels, Nature, Vol. 367, No. 6463, 1994, p. 546-548

Degryse, F., Smolders, E., Zhang, H. & Davison, W. 2009, "Predicting availability of mineral elements to plants with the DGT technique: A review of experimental data and interpretation by modelling", Environmental Chemistry, vol. 6, no. 3, pp. 198-218.

Dodson S.I. (2005), Aquatic ecosystems: Chemical cycles, In: Introduction to Limnology, Mc Graw Hill, Higher education, Boston, 239- 245.

Eggen, T, 2013, VRI project for Rogaland County: Characterization of phosphorus in manure – a literature study and knowledge transfer to innovation, Bioforsk Vest Særheim, Klepp, pers. comm.

Fatin-Rouge,N, Milon, A., Buffle, J, Goulet R.R., Andre´ Tessier, A. (2003), Diffusion and Partitioning of Solutes in Agarose Hydrogels: The Relative Influence of Electrostatic and Specific Interactions, J. Phys. Chem. B 2003, 107, 12126-12137

Frigon RP, Leypoldt JK, Uyeji S, Henderson LW (1983): Disparity between Stokes radii of dextrans and proteins as determined by retention volume in gel permeation chromatography. Anal Chem 55:1349–1354

Garmo, Ø, A., Røyset, O., Steinnes, E., Flaten, T, P.(2003).Performance study of diffusive gradients in thin films for 55 elements. Analytical Chemistry, 75, 3573-3580.

Garmo, Ø. A., Naqvi, K., Røyset, O., and Steinnes, E. (2006) Estimation of diffusive boundary layer thickness in studies involving diffusive gradients in thin films (DGT), Analytical and Bioanalytical Chemistry 386, 2233–2237.

Ged EC, Boyer TH, (2013), Molecular weight distribution of phosphorus fraction of aquatic dissolved organic matter, Chemosphere 91, , 921–927.

http://www.mn.uio.no/kjemi/english/research/projects/EUTROPIA/

http://www.mn.uio.no/kjemi/english/research/projects/sinotropia

ISO 15681-1 : 2003, Water quality -- Determination of orthophosphate and total phosphorus contents by flow analysis (FIA and CFA) -- Part 1: Method by flow injection Analysis (FIA). International Standardisation Organisation, www.iso.org.

ISO 15681-2 : 2003, Water quality -- Determination of orthophosphate and total phosphorus contents by flow analysis (FIA and CFA) -- Part 2: Method by continuous flow analysis (CFA).). International Standardisation Organisation, www.iso.org.

Konieczka, P, Namiesnik, J, 2009, Quality Assurance and Quality Control in the Analytcial Laboratory, A practical approach, CRC Press, Taylor and Francis, Boca Raton, FL, US

Kortelainen, P., Mattsson, T., Laubel, A., Evans, D., Cauwet, G. & Räike, A. 2004. Sources of dissolved organic matter from land. In: Søndergaard, M. & Thomas, D. N. (eds.) Dissolved Organic Matter (DOM) in Aquatic Ecosystems: A Study of European Catchments and Coastal Waters. The Domaine project.

Krogstad, T., Øgaard, A., and Skarbøvik, E. (2013) Laboratorieanalyser av suspendert stoff, fosfor og nitrogen i turbide vannprøver - usikkerhet og metodeutfordringer, Vann, 2, 239-248, 2013.

Lead, JR, Starchev, K, Wilkinson, KJ, (2003) Diffusion Coefficients of Humic Substances in Agarose Gel and in Water, Environ. Sci. Technol. 2003, 37, 482-487,

Li, Yuan-Hui, Gregory, S., 1974, Diffusion of ions sin seawater and in deep-sea sediments, Geochimica and Cosmochimica Acta, 1974, 28, 703-714.

Logan B E (2012) Environmental Transport Processes, Wiley-Blackwell; 2nd Ed. 482 pp, ISBN-10: 0470619597.

Logan, B. E., (2014) Molecular and Colloidal Sizes of DOM, Department of Civil & Environmental Engineering, The Pennsylvania State University. Available at the web: http://www.engr.psu.edu/ce/enve/576/576-Molecular-size-spectra-2011.pdf, downloaded at 20140121.

Mason, S., Hamon, R., Nolan, A., Zhang, H. & Davison, W. 2005, "Performance of a mixed binding layer for measuring anions and cations in a single assay using the diffusive gradients in thin films technique", Analytical Chemistry, vol. 77, no. 19, pp. 6339-6346.

Mason, S., Hamon, R., Zhang, H. & Anderson, J. 2008, "Investigating chemical constraints to the measurement of phosphorus in soils using diffusive gradients in thin films (DGT) and resin methods", Talanta, vol. 74, no. 4, pp. 779-787.

Mason, S., McNeill, A., McLaughlin, M.J. & Zhang, H. 2010, "Prediction of wheat response to an application of phosphorus under field conditions using diffusive gradients in thin-films (DGT) and extraction methods", Plant and Soil, vol. 337, no. 1, pp. 243-258.

Moorleghem, CV, Six L, Degryse F, Smolders E, Merckx; R (2013) Effect of Organic P Forms and P Present in Inorganic Colloids on the Determination of Dissolved P in Environmental Samples by the Diffusive Gradient in Thin Films Technique, Ion Chromatography, and Colorimetry, Anal. Chem. 2011, 83, 5317–5323, (dx.doi.org/10.1021/ac200748e |)

Murphy, J.; Riley, J. P. 1962, A modified single solution method for the determination of phosphate in natural waters. Analytica Chimica Acta 1962, 27, 31-36.

Norsk Standard methods 1984, NS4724:1985, Vannundersøkelse - Bestemmelse av fosfat, (Water quality - Determination of phosphorus - Ammonium molybdate spectrometric method), , Norsk Standardiserings Forbund/Standard Norge,

Norsk Standard methods 1984, NS4725:1984, Vannundersøkelse - Bestemmelse av totalfosfor - Oppslutning med peroksodisulfat (Water analysis - Determination of total phosphorus - Digestion by peroxodisulphate), Norsk Standardiserings Forbund/Standard Norge.

NS-EN-ISO 6878:2004, Water quality - Determination of phosphorus - Ammonium molybdate spectrometric method (ISO 6878:2004), Norsk Standardiserings Forbund/Standard Norge.

Orderud, G. I., & Vogt, R. D. (2013). Trans-disciplinarity required in understanding, predicting and dealing with water eutrophication. TSDW International Journal of Sustainable Development & World Ecology. doi: http://dx.doi.org/10.1080/13504509.2013.814605

Panther, J.G., Teasdale, P.R., Bennett, W.W., Welsh, D.T. & Zhao, H. 2010, "Titanium dioxide-based DGT technique for in situ measurement of dissolved reactive phosphorus in fresh and marine waters", Environmental Science and Technology, vol. 44, no. 24, pp. 9419-9424.

Panther, J.G., Teasdale, P.R., Bennett, W.W., Welsh, D.T. & Zhao, H. 2011, "Comparing dissolved reactive phosphorus measured by DGT with ferrihydrite and titanium dioxide adsorbents: Anionic interferences, adsorbent capacity and deployment time", Analytica Chimica Acta, vol. 698, no. 1-2, pp. 20-26.

Panther, J.G., Stewart, R.R., Teasdale, P.R., Bennett, W.W., Welsh, D.T. & Zhao, H. 2013, "Titanium dioxide-based DGT for measuring dissolved As(V), V(V), Sb(V), Mo(VI) and W(VI) in water", Talanta, vol. 105, pp. 80-86.

Pascal L.R. , van der Veeken, P.L.R, Pinheiro, J.P. , van Leeuwen, H.P.(2008) Metal Speciation by DGT/DET in Colloidal Complex Systems, Environ. Sci. Technol., 2008, 42 (23), 8835-8840.

Perdue EM, Ritchie JD, (2003) Dissolved Organic Matter in Freshwater, In: Turekian, K, Holland, HH (Editors), Treatise on Geochemistry, Ten Volume Set, Volume 5 pp. 273-318, ISBN 0-08044340-0, Elsevier Ltd,

Perdue, E. M. 2009. Natural Organic matter. In: Likens, G. E. (ed.) Reference Module in Earth Systems and Environmental Sciences, from Encyclopedia of Inland Waters. Elsevier, pp 805-819.

Polson, A. (1950) Some aspects of diffusion in solution and a definition of a colloidal particle. J. Phys. Colloid Chem. 54:649-652.

Price H.L, Teasdale, P.R., Jolley, D.F. (2013), An evaluation of ferrihydrite- and Metsorb-DGT techniques for measuring oxyanion species (As, Se, V, P): Effective capacity,competition and diffusion coefficients, Analytica Chimica Acta 803 (2013) 56– 65.

Qualls, R. G. & Richardson, C. J. 2003. Factors controlling concentration, export, and decomposition of dissolved organic nutrients in the Everglades of Florida. Biogeochemistry, 62, 197-229.

Røyset, O.; Bjerke, E.; Eich-Greatorex, S.; Sogn, T. A.; Almås, Å. R. Simultaneuos sampling of phosphate, arsenate and selenate in water by Diffusive Gradients in Thin Films (DGT); NIVA, Norwegian Institute for Water Research: 2004; p 17.

Røyset, O, Vogt, R.D, (2014) Fractionation of phosphorus compounds in water, NIVA report 6622-2014, Norwegian Institute for Water Research, NIVA , Oslo

Røyset, O, (2013a) Advances in phosphorous analysis, RCN Eutropia Final Project Conference 30-31. May 2013 CIENS, Oslo. Available at the web: http://www.mn.uio.no/kjemi/english/research/projects/eutropia/presentations/project-presentations-in-2013/final-project-conference/royset.pdf

Røyset, O, (2013b) Fremtidens overvåking av fosfor, RCN Eutropia Final Project Conference 30-31. May 2013, CIENS, Oslo. Available at the web: http://www.mn.uio.no/kjemi/english/research/projects/eutropia/presentations/project-presentations-in-2013/brukernes-dag/royset_eutropia_workshop-20130531_brukermote.pdf

Røyset, O., Garmo, O.A. (2002), Studies of D for heavy metal cations in the CGA membrane, unpublished results.

Skarbøvik, E., & Bechmann, M. (2010). Some Characteristics of the Vansjø-Hobøl (Morsa) Catchment, BioForsk report (Vol. 5, pp. 46). BioForsk, Ås, N1432 ÅS.

Skarbøvik, E., S., H., & Bechmann, M. (2013). Overvåking Vansjø/Morsa 2011-2012. Resultater fra overvåking, BioForsk, Ås, N1432 ÅS.

Skarbøvik, E, Haande, S, 2012, Overvåkning Vansjø/Morsa 2010-2011. Resultater fra overvåking av innsjøer, elver og bekker i perioden oktober 2010 - oktober 2011, Bioforsk report (Vol 7, nr 44-2012), 121 pp, BioForsk, Ås, N1432 ÅS.

Six, L., Smolders, E. , Merckx, R. 2012, "In contrast with established soil P tests, the diffusive gradients in thin films (DGT) technique accesses the same phosphorus (P) pool as plants in strongly weathered tropical soils.", Communications in agricultural and applied biological sciences, vol. 77, no. 1, pp. 127-131.

Sogn, T.A., Eich-Greatorex, S., Røyset, O., Øgaard, A.F., Almås, Å.R. 2008, "Use of diffusive gradients in thin films to predict potentially bioavailable selenium in soil", Communications in Soil Science and Plant Analysis, vol. 39, no. 3-4, pp. 587-602.

Turner, B:L:, Frossard, E. Baldwin D.S. (Editors), (2004) , Organic Phosphorus in the Environment, 432 pp. Cabi Publishers, UK

Tandy, S., Mundus, S., Yngvesson, J., de Bang, T.C., Lombi, E., Schjoerring, J.K. & Husted, S. 2011, "The use of DGT for prediction of plant available copper, zinc and phosphorus in agricultural soils", Plant and Soil, vol. 346, no. 1, pp. 167-180.

Vogt, R. D. (2012). Water quality in a changing environment. Public Service Review: European Union 2012, 23, 386-387.

Vogt (2014), personal communication on the interpretation of Mw data from Perdue et al. (2009)

Vogt. R (2013) Reduction in Acid rain and Climate change are disguising abatement effects, EUTROPIA Final conference, 30. May 2013, http://www.mn.uio.no/kjemi/english/research/projects/eutropia/presentations/project-presentations-in-2013/final-project-conference/vogt-final-eutropia-conference-130525-for-pdf.pdf

Zhang, H., Davison, W., Gadi, R. & Kobayashi, T. 1998, In situ measurement of dissolved phosphorus in natural waters using DGT, Analytica Chimica Acta, vol. 370, no. 1, pp. 29-38.

Zhang, H, Davison W, (1999) Diffusional characteristics of hydrogels used in DGT and DET techniques, Analytica Chimica Acta 398 (1999) 329–340.

Zhang, H. DGT - for measurements in water, soil and sediments; DGT Research Ltd: Lancaster, 2005, web version.

Zhang, Y., Mason, S., McNeill, A. & McLaughlin, M.J. 2013, "Optimization of the diffusive gradients in thin films (DGT) method for simultaneous assay of potassium and plant-available phosphorus in soils", Talanta,

Zhu Y, Wu F, He Z, Guo J, Qu X, Xie F, Giesy FP, Liao H, Guo F (2013), Characterization of Organic Phosphorus in Lake Sediments by Sequential Fractionation and Enzymatic Hydrolysis , ES&T, 2013,47, 7679-7687.

Warnken, K. W., Zhang, H., and Davison, W. (2006) Accuracy of the diffusive gradients in thin-films technique: Diffusive boundary layer and effective sampling area considerations, Analytical Chemistry 78, 3780–3787).

Worsfold PJ, Monbeta,P, Tappina, AD, Fitzsimonsa, MF, Stiles, DA, McKelvie ID, (2008) , Characterisation and quantification of organic phosphorus and organic nitrogen components in aquatic systems: A Review, Analytica Chimica Acta 624 (2008) 37–58.

Appendix A. Data fordiffusion coefficients

Table 27 of this appendix contains detailed D values in the range 10 to 1000 000 Da based on the different equations in Table 12.

Table 26.
Diffusion coefficient values (10^{-6} cm^2/sec) obtained from the predictions by the equation in Table 12. The Jones 2014 equation is based on Buffle1988, with a correction for porevolume of 4% according to Archies law.

Molecular weight	Molecular Diameter Logan 2012	BUFFLE 2007	BECKET 1987 modified	POLSON 1950	FRIGON 1983	Jones 2014 (Buffle 1988x 0.96)	BUFFLE 1988	Average From all equations
Da								
10	0,23	13,28	22,7	12,82	23,85	14,82	15,44	17,93
25		9,82	15,42	9,47	15,51	10,96	11,41	12,55
50		7,81	11,51	7,54	11,20	8,72	9,08	9,61
100	0,62	6,21	8,59	5,99	8,08	6,93	7,22	7,36
200		4,94	6,41	4,77	5,84	5,52	5,74	5,66
347		4,12	5,08	3,98	4,50	4,60	4,79	4,59
500		3,65	4,36	3,52	3,79	4,08	4,24	4,00
736		3,22	3,70	3,10	3,16	3,59	3,74	3,46
1000	1,4	2,91	3,25	2,80	2,74	3,24	3,38	3,08
2000		2,31	2,43	2,23	1,98	2,58	2,69	2,38
2400		2,18	2,25	2,10	1,81	2,43	2,53	2,22
3000		2,02	2,05	1,95	1,63	2,26	2,35	2,05
5000		1,71	1,65	1,65	1,29	1,91	1,99	1,70
6000		1,61	1,53	1,55	1,18	1,80	1,87	1,58
6300		1,58	1,50	1,53	1,15	1,77	1,84	1,56
10000	2,8	1,36	1,23	1,31	0,93	1,52	1,58	1,31
20000		1,08	0,92	1,04	0,67	1,21	1,26	1,02
23000		1,03	0,87	1,00	0,63	1,15	1,20	0,97
50000		0,80	0,62	0,77	0,44	0,89	0,93	0,73
100000	6,2	0,64	0,47	0,61	0,31	0,71	0,74	0,57
500000		0,37	0,24	0,36	0,15	0,42	0,43	0,32
1000000	13,2	0,30	0,18	0,29	0,11	0,33	0,35	0,25

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