

In situ Phosphate Measurement Using Fe₃O₄/Chelex®100-Agarose-Oxalic Acid Hydrogel in Diffusive Gradient in Thin Films

Layta Dinira, Barlah Rumhayati*, Diah Mardiana, Ulfa Andayani

Departemen of Chemistry, Faculty of Mathematics and Natural Sciences, Brawijaya University, Malang 65142, Indonesia

*Corresponding author email: rumhayati_barlah@ub.ac.id

Received January 17, 2024; Accepted May 16, 2024; Available online July 20, 2024

ABSTRACT. This study would be the first to develop a novel combination of Chelex®100 and Fe₃O₄ as a mixed binding gel for in situ phosphate measurement employing Diffusive Gradient in Thin Films (DGT). Fe₃O₄ and Chelex®100 were utilized as binding agents within a single binding layer for phosphate measurements in laboratory and natural water settings. The synthesized Fe₃O₄ was identified as magnetite and consisted of micro-sized particles. The pore size of the mixed binding gel ranged from 22.39 to 112.5 µm. Incorporating Chelex®100 with Fe₃O₄ in oxalic acid cross-linked agarose did not diminish phosphate adsorption efficiency. As adsorption time and phosphate concentration in solution increased, the quantity of adsorbed phosphate in the mixed binding gel also increased. Optimal phosphate elution was achieved using a basic solution, with a phosphate elution efficiency of $95.3 \pm 0.4\%$ observed with 0.4 M NaOH. The diffusion coefficient measured using the mixed binding gel was 1.08 times greater than that of polyacrylamide cross-linked with an agarose derivative (APA) gel, typically employed in the DGT technique. Phosphate concentration measurement with Fe₃O₄/Chelex®100-agarose oxalic acid in a DGT passive sampler at pH 4-8 yielded values twice those in bulk solution. Similar results were obtained when measuring phosphate in a 0.01 – 0.1 M NaNO₃ solution. A T-test showed that the phosphate concentration obtained from mixed binding layer-DGT as an alternate passive sampler was not significantly different from grab sampling. This study underscores the suitability of Fe₃O₄-Chelex®100 impregnated in agarose-oxalic acid gel for monitoring phosphate in natural water via DGT.

Keywords: Agarose, diffusive gradient in thin films, mixed binding gel Fe₃O₄, oxalic acid.

INTRODUCTION

Phosphate is essential for plant growth and development (Y. Huang et al., 2020). Phosphorus, in the form of phosphate, is used as a fertilizer. Excess phosphate in the environment goes directly into the water, drastically increasing its concentration; this can cause eutrophication, which can reduce water quality and the number of organisms living in the water (Franco et al., 2017; Wurtsbaugh et al., 2019). Therefore, monitoring phosphate concentration in water is vital.

Phosphate measurements in water can be done using in situ passive sampling techniques. Passive sampling allows analytes to diffuse from the sample to a receiving medium with a high affinity for the target analyte (Shakallis et al., 2022). The media receiver or passive sampler effectively monitors contamination in environmental samples, including water, soil, and air (Booij et al., 2016). Unlike spot or grab sampling, commonly used for monitoring, passive samplers can be left in sampling locations for an extended time (hours, days, weeks, or months). Grab sampling can only determine analyte concentration at the time of sampling, whereas passive sampling provides more

information through the average analyte concentration over time (Taylor et al., 2020). Furthermore, removing interference in grab sampling requires additional procedures that could physically or chemically harm the sample.

The diffusive gradient in thin films (DGT), invented 30 years ago, is a passive sampling commonly used on environmental samples such as water, soil, and sediment. It is suitable for environmental monitoring due to its practicality and ease of use for in situ deployment, applicability to various environmental matrices, and ability to measure a wide range of target chemicals (Marrugo-Madrid et al., 2021). Recent water monitoring studies used DGT to detect labile methylmercury (Bajagain et al., 2024), polycyclic aromatic hydrocarbons (PAH) (S. Ren et al., 2024), and pharmaceuticals (Cao et al., 2024). DGT can also monitor trace metals and oxyanions in sediments (Lin & Pan, 2023).

DGT passive samplers comprise a binding layer that irreversibly accumulates analytes, a diffusive layer that allows analytes to pass through, and a filter membrane that protects the diffusive layer against particles (C. Li et al., 2019). DGT passive sampling

removes interfering compounds in analyte measurements due to analytes' different affinities and interference with the binding agent. DGT enables in situ preconcentration as analytes accumulate, making the measurement more sensitive. Binding gels continue to be developed to obtain reliable and robust DGT in challenging environments (C. Zhang et al., 2014).

The hydrogels typically used in DGT are open-pored polyacrylamide gels containing an agarose-based cross-linker with pore sizes >5 nm (H. M. Pouran et al., 2014; H. Zhang & Davison, 1999). The acrylamide monomer, however, is categorized as a probable human carcinogen by the International Agency for Research on Cancer. Therefore, it is possible to use agarose gel with pore sizes >20 nm (Wang et al., 2016); to prevent binding gels from absorbing large molecules such as colloids and complex compounds containing organic ligands (Shiva et al., 2015), agarose pore sizes are reduced by cross-linking with oxalic acid.

DGT is mostly used to adsorb cations or anions only. The binding agent commonly used for adsorbing cations is Chelex®100, while ferrihydrite, titanium dioxide-based adsorbent (Metsorb), Zr oxide, and AgI are used to adsorb anions (Sun et al., 2015). Fe_3O_4 was offered as an alternative binding agent for phosphate measurements using DGT. Fe_3O_4 -DGT capacity is approximately $105 \mu\text{g}$ phosphate per passive sampler (Y. Zhang et al., 2018).

A mixed-binding gel has been developed to measure cations and anions simultaneously, a single binding layer containing a mixture of binding agents. A mixture of Chelex®100 and ferrihydrite is developed as a single binding gel for measuring As, Cd, Cu, Zn, Pb, and Mo, Chelex®100 and Metsorb for V, As, Mo, Sb, W, P, etc., and Chelex®100 and ZrO for Fe, As, and P (Sun et al., 2015). Wang et al. (2017) used Chelex®100 and ZrO to measure eight cations and eight anions.

Mixed binding layers that adsorb multiple analytes save preparation, deployment, analysis time, and materials compared to DGT passive samplers designed for a single analyte. Nevertheless, using a mixed binding layer for many analytes may increase competition with other ions and decrease the binding capacity for individual analytes (J. Huang et al., 2017). Thus, using mixed binding layer-DGT for long-term deployment will be limited (Feng et al., 2018), and using Fe_3O_4 as a new high-capacity binding agent for phosphate is required. Furthermore, the specific behavior of the mixed binding gel (Chelex- Fe_3O_4) to measure phosphate must be evaluated.

This study introduces Fe_3O_4 /Chelex®100 impregnated in oxalic acid cross-linked agarose gel for measuring phosphate in water using DGT. Laboratory experiments were conducted to test the gel's adsorption and elution capacity. Binding layers

were assembled to form DGT samplers to measure phosphate in laboratory conditions and natural water.

EXPERIMENTAL SECTION

Materials and Apparatus

Deionized water was used to prepare all solutions of KH_2PO_4 , NaNO_3 , NaOH , NH_3 , H_2SO_4 , HNO_3 , and HCl provided by Merck (analytical grade). KH_2PO_4 powders, NaNO_3 powders, NaOH pellets, and concentrated solutions, such as NH_3 25%, H_2SO_4 98%, HNO_3 65%, and HCl 32%, were utilized to make the solutions in this study. KH_2PO_4 was used to make phosphate solution, and NaNO_3 was used to make 0.01 - 0.1 M NaNO_3 solution. NaOH and concentrated NH_3 were used to make basic solutions, while concentrated H_2SO_4 , HNO_3 , and HCl were used to make acidic solutions. The pH of the solutions was adjusted using acid and base solutions.

Agarose powder (99% purity, Himedia, India) and oxalic acid powder (99% purity, Merck, Germany) were used for diffusive gels. Both materials were also used with Chelex®100 (Bio-Rad, 200-400 mesh, sodium form) to make binding gels. Fe_3O_4 was synthesized from $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, and $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ was obtained from Merck.

All DGT passive samplers used in this study were solution deployments purchased from DGT Research Ltd. A Nitrocellulose membrane ($0.45 \mu\text{m}$) from Merck was used as a membrane filter. Two separated glasses with plastic spacers 0.08 cm thick were used to make diffusive and binding gels.

Gel Preparation

Agarose-based diffusion gels (Δg) 0.08 cm thick and 2.5 cm diameter were prepared following Eismann et al. (2023). Oxalic acid (10% w/w of agarose) was added to a solution containing 1.5% agarose at 60°C . Crosslinking was performed in an oven for 15 minutes at 40°C . The gel discs were stored in 0.1 M NaNO_3 .

Fe_3O_4 , as the binding agent for phosphate, was synthesized using the coprecipitation method following Wulandari et al. (2018), except that the weights of $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ and $\text{FeCl}_3 \cdot 7\text{H}_2\text{O}$ were adjusted to 2.78 g and 5.41 g, respectively. Fe_3O_4 was characterized by Scanning Electron Microscopy (Hitachi TM3000) and X-ray diffraction (PANalytical X'Pert3 Powder). The mixed binding gels containing a mixture of Fe_3O_4 and Chelex®100 were incorporated in 10 mL agarose-oxalic acid gel solution with a total binding agent of 2 g. The gel was immersed in deionized water for at least 24 hours with three water replacements before being stored in deionized water. The binding gel thickness was 0.08 cm.

Phosphate Uptake Efficiencies

Mass ratios of Chelex®100 to Fe_3O_4 , adsorption time, and phosphate concentration in solutions were observed to study the phosphate uptake efficiencies. To analyze the effect of binding agent quantity on phosphate uptake efficiencies, triplicate binding gels

(2.5 cm diameter) with five ratios of Chelex®100 to Fe₃O₄ (w:w) were immersed in 15 mL of 2 mg/L phosphate solution. The solutions were shaken at 100 rpm for 24 hours. The gel containing optimum mass ratios of Fe₃O₄ and Chelex®100 was tested for the binding kinetics. Triplicate gels were placed in an Erlenmeyer flask containing 15 mL of phosphate (2 mg/L) at 20 and 40 minutes and 1, 2, 4, and 24 hours. To observe phosphate binding affinity onto the mixed binding gel, triplicate mass ratios gels were placed in 15 mL of solution containing 0.4, 0.8, 1.2, 1.6, 2, and 2.4 mg phosphate/L, shaken for 4 hours at room temperature. At each experiment, the gel discs and filtrate were separated, and the initial phosphate concentration in the filtrate was determined using a UV-Vis spectrophotometer with the Stannous Chloride method following conversion to the initial phosphate mass (M_i) and the mass of phosphate in the remaining solution after the experiment (M_f). The phosphate uptake or adsorption factor (f_a , %) was calculated following Equation (1).

$$f_a (\%) = (M_i - M_f) / M_i \times 100\% \quad (1)$$

Elution Efficiencies

Fe₃O₄/Chelex®100-agarose-oxalic acid gel discs (Fe₃O₄:Chelex®100 0.75:1.25 g) were placed in 15 mL of 0.8 mg/L phosphate solution and shaken for four hours at 100 rpm. The mixed binding gels were retrieved, rinsed with deionized water, immersed in 15 mL of eluent, and eluted at 100 rpm for 24 hours. The eluent included various acid solutions and different NaOH concentrations. The influence of eluent volume was determined using 15, 20, and 25 mL of 0.4 M NaOH. Elution kinetics were established by eluting mixed binding gels in 20 mL of 0.4 M NaOH at 20 and 40 minutes and 1, 2, 4, and 24 hours. The phosphate adsorption factor (%) was calculated following Equation (1), while the elution factor (f_e , %) was calculated following Equation (2) after measuring the phosphate concentration in the eluent solution converted into mass (M_e).

$$f_e (\%) = M_e / (M_i - M_f) \times 100\% \quad (2)$$

DGT Assembly

DGT samplers were assembled using a DGT holder for solutions deployment (DGT Research Ltd.) with a 2-cm-diameter exposure window (geometric sampling area of 3.14 cm²). On the holder's bottom, a Fe₃O₄/Chelex®100-agarose-oxalic acid gel was placed, followed by an agarose-oxalic acid diffusion gel and a 0.45 m nitrocellulose membrane atop the diffusion gel. DGT samplers were used in triplicate for every experiment. Regular monitoring of the solution pH and temperatures was conducted. Before deploying DGT samplers to the solution, the solution's pH and temperature were determined and recorded. During deployment, pH and temperature were measured and recorded every hour until the deployment time was up.

Diffusion Coefficient of Phosphate

The phosphate diffusion coefficient was determined from the mass accumulation over time (DGT time-series deployment). Six assembled DGT samplers, as mentioned earlier, were deployed in 2 L of 0.8 mg/L phosphate solution stirred at 200 rpm. The diffusion gel was agarose-oxalic acid stored in deionized water. One DGT passive sampler was retrieved at 1, 2, 3, 4, 5, and 6 hours. The binding gel was rinsed with deionized water, and the accumulated phosphate was eluted with 20 mL of 0.4 M NaOH at 100 rpm for one hour. The binding gel and filtrate were separated. The phosphate concentration in the filtrate was measured for absorbance using a UV-Vis spectrophotometer using the Stannous Chloride method. The experiment was performed three times.

The filtrate concentrations were converted into mass units (M , ng) using Equation (3). C_e is the analyte concentration in the eluted solution, V_g is the gel volume, V_e is the filtrate volume, and f_e is the elution factor. A linear regression was created for the phosphate mass accumulated in the binding gel.

$$M = C_e (V_g + V_e) / f_e \quad (3)$$

The phosphate diffusion coefficient (D , cm²/s) is calculated following Equation (4). *Slope* is the slope of the curve of the relationship between diffusion time and phosphate mass absorbed by the binding gel, the diffusion layer thickness Δg (0.08 cm), the diffusion area A (3.14 cm²), and the solution concentration C_{sol} (ng/mL). The temperature correction for the diffusion coefficient to 25°C follows the Stokes-Einstein equation (Sun et al., 2015). The afore-mentioned procedure was repeated for agarose-oxalic acid stored in 0.1 M NaNO₃.

$$D = (\text{slope} \Delta g) / (C_{sol} A) \quad (4)$$

Determining DGT Blank and Method Detection Limits

The blank for phosphate was measured by immersing three replicates of DGT samplers containing Fe₃O₄/Chelex®100-agarose oxalic acid gel into a 2 L well-stirred solution pH 4 – 8 and 0.01 and 0.1 M NaNO₃ for four hours. The mixed binding gels were retrieved, rinsed with deionized water, and eluted using 20 mL of 0.4 M NaOH for one hour at 100 rpm. The gel and filtrate were separated, and the phosphate concentration in the filtrate was measured using a UV-visible spectrophotometer.

The filtrate concentration value was then converted into mass units (ng) using Equation (3). The method detection limit (MDL) was calculated three times the standard deviations of the blanks. MDL was converted to DGT concentrations (C_{DGT}), as in Equation (5), using the diffusion thickness (Δg) of 0.08 cm, gel contact area (A) of 3.14 cm², deployment time (t) of four hours, and the phosphate diffusion coefficient of agarose-oxalic acid stored in 0.1 M NaNO₃ corrected to the experiment's temperature.

$$C_{DGT} = (M \Delta g) / (D A t) \quad (5)$$

Effect of pH and Ionic Strength on DGT Uptake

To investigate the effect of pH on the accumulation of phosphate by Fe_3O_4 /Chelex®100-agarose-oxalic acid-DGT, three replicates of DGT samplers containing mixed binding gel were immersed in 2 L well-stirred phosphate solution at pH 4 – 8; the pH was adjusted using diluted HCl and NaOH. The pH of the solutions was measured before and during deployment to monitor and maintain in case of dramatic changes during this experiment. During deployment, the pH of the solutions was measured every hour until the deployment time was up. The effect of ionic strength on mixed binding gels-DGT performance was tested by deploying the three replicates in 2 L phosphate solution at 0.01 and 0.1 M NaNO_3 . The phosphate concentration was 0.8 mg/L, and the deployment time of DGT was set at four hours for both experiments. The mixed binding gels were eluted using 20 mL of 0.4 M NaOH for one hour at 100 rpm, and the phosphate concentration in the filtrate was measured. Equations 3 and 6 were used to convert the phosphate concentration in the filtrate to mass and C_{DGT} , respectively. The average phosphate concentration in the solution (before and after the experiment) was measured and stated as C_{sol} to determine the relationship between C_{DGT} and C_{sol} .

Application of DGT Samplers in Natural Water

The performance of Fe_3O_4 /Chelex®100-agarose-oxalic acid as the binding gel was assessed by deploying the DGT samplers in natural freshwater collected from Arboretum, Sumber Brantas, Bumiaji District, Batu City. The sampling site was Titik Nol Sungai Brantas (7°45'14"S 112°31'35"E). Natural freshwater sampling was done at three points by scooping the water at a depth of 10 cm using a dipper, 1.5 L of water from each point, and then placing it into HDPE bottles. The HDPE bottles containing natural freshwater were put into a cooler box containing ice gel packs and taken to the laboratory. This sampling was conducted once (10:00 A.M. Western Indonesian Time / UTC+7).

The phosphate concentration in natural freshwater was measured immediately upon arrival at the laboratory. Triplicate DGT samplers were immersed in 2 L well-stirred natural freshwater. After four hours, the DGT samplers were retrieved, and the mixed binding gels were rinsed with deionized water. The phosphate concentration in the filtrate of natural freshwater was eluted and detected using the method described earlier, which was then converted to mass and C_{DGT} . The phosphate concentration before and after DGT deployment was also measured. The average phosphate concentrations were expressed as C_{sol} .

To obtain a more accurate C_{DGT} of phosphate in natural water, the C_{DGT} of phosphate in natural water was plotted against a C_{DGT} calibration curve from a phosphate standard solution with a matrix similar to natural water (synthetic soft water). The mixed binding gel-DGT was placed in 2 L well-stirred synthetic soft

water containing 0.2, 0.4, 0.6, 0.8, and 1 mg/L of phosphate solution. The synthetic soft water with various phosphate concentrations was prepared following Smith, Davison, & Hamilton-Taylor (2002). Like the previous procedure, the DGT sampler was retrieved, the phosphate was eluted from mixed binding gels, and the mass to C_{DGT} was calculated. A calibration curve was constructed between the phosphate standard solution concentration and the C_{DGT} . The phosphate concentration of natural water derived from plotting the C_{DGT} of natural water to the calibration curve was compared to the phosphate concentration measured directly from the natural freshwater (C_{sol}) to determine the accuracy of the mixed binding gel-DGT.

Phosphate Analysis

The Stannous Chloride method, which yielded a blue solution, was used to determine the phosphate in the samples. To 4 mL of sample solution, 0.16 mL of ammonium molybdate solution and 20 μL of SnCl_2 reagent were added. The solutions were left for ± 10 minutes. A UV-Vis spectrophotometer (Thermo Scientific Genesys 10S) was used to measure the absorbance of the solution at 695 nm.

RESULTS AND DISCUSSION

Synthesis of Fe_3O_4

The scanning electron microscopy (SEM) picture depicting Fe_3O_4 powder is presented in **Figure 1a**. Fe_3O_4 with unequal sizes and uneven shapes were visible in SEM. The size of the particles on the surface of Fe_3O_4 ranges from 0.267 to 1.25 μm . In addition, the Particle Size Analyzer test confirmed the average diameter of Fe_3O_4 of 5.620 μm , indicating microscale Fe_3O_4 formation. The strong magnetism and high stirring speed can lead to the aggregation of Fe_3O_4 nanoparticles, resulting in the formation of microscale particles (Fadli et al., 2019; Hou et al., 2020).

Synthesized Fe_3O_4 was solid and black. The presence of black suggested that the particles produced were magnetite (Lenders et al., 2017). **Figure 1b** shows the XRD diffractogram of the Fe_3O_4 . The peaks observed in the X-ray diffraction pattern of magnetite are located at specific angles (2θ) corresponding to 30°, 35°, 43°, 54°, 57°, and 63°. These peaks indicated the (220), (311), (400), (422), (511), and (440) planes (De Rezende Bonesio & Pissetti, 2020).

Binding Layer Capacity

Figure 2a shows that the phosphate adsorption efficiency in all compositions of Chelex®100 and Fe_3O_4 was not different ($\approx 98 - 99\%$). The efficacy of phosphate adsorption was significantly affected by Fe_3O_4 . Phosphate uptake by the Fe_3O_4 single-binding gel was 99%. Chelex®100, impregnated in oxalic acid cross-linked agarose gel, could adsorb phosphate ($12.7 \pm 2.6\%$). Nevertheless, at neutral and alkaline pH, the iminodiacetic acid group ($\text{COOHCH}_2\text{-NH-COOHCH}_2$) of Chelex®100 is negatively charged,

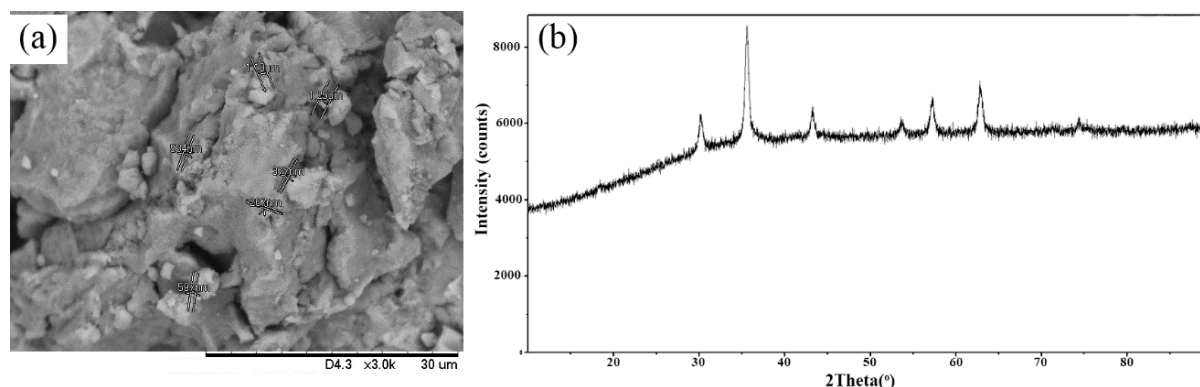


Figure 1. (a) SEM image at 3000x magnification and (b) XRD pattern of Fe_3O_4

which can cause electrostatic repulsion towards phosphate (H. Pouran et al., 2020; Smolíková et al., 2022). Phosphate trapped in the gel matrix may cause phosphate uptake by single binding gel Chelex®100. Hence, the coexistence of Chelex®100 and Fe_3O_4 within oxalic acid cross-linked agarose gel did not reduce the percentage of phosphate adsorption.

The mixed binding gels containing Chelex®100: Fe_3O_4 1:3 and 3:5 had the highest phosphate adsorption efficiency (99.5%). A statistical test was conducted to determine whether the phosphate adsorption efficiency differed significantly at each mass ratio of binding agents. The statistical test showed a significance value of $0.02 < \alpha = 0.05$. This result demonstrated that the phosphate adsorption efficiencies at a mass ratio of 1:7, 1:3, 3:5, 1:1, and 5:3 were significantly different.

A balanced mix of cation and anion-binding agents is necessary when DGT samplers are used for simultaneous analysis. Consequently, a mixture consisting of Chelex®100 and Fe_3O_4 in a ratio of 3:5 was the best option. It is also necessary to use the optimum composition of the binding agent to maintain the strength and thickness of the binding gel to ensure a good performance of analyte measurements using DGT (Wang et al., 2019). **Figure 3** shows the SEM image of Fe_3O_4 and Chelex®100 impregnated in oxalic acid cross-linked agarose with a composition of 3:5. The gel has an irregular pore size distribution, from 22.39 to 112.5 μm .

Figure 2b illustrates the mass of phosphate adsorbed by Chelex®100- Fe_3O_4 -agarose gel cross-linked with oxalic acid at a specific time. In the first 60 minutes, phosphate adsorption efficiency increased significantly. A similar result was observed when ZrO-Chelex®100 binding gel was utilized (Xu et al., 2013). Phosphate adsorption increased linearly for the first 60 minutes, then slowed to relatively constant until 120 minutes. The phosphate adsorption efficiency exceeded 90% and reached >98% in 120 minutes when ZrO-Chelex®100-Agl gel was used (Wang et al., 2019). Nonetheless, within four hours, the gel demonstrated good phosphate adsorption ($83.1 \pm 0.2\%$). Adsorption equilibrium was reached within four

hours and continued until 24 hours (**Figure 2b**). Therefore, four hours of adsorption time was employed in the following step.

Figure 2c depicts that the mass of phosphate adsorbed by the mixed binding gel increases linearly to the phosphate concentration in the solution. A similar result was demonstrated by Kreuzeder, Santner, Prohaska, & Wenzel (2013) using zirconium hydroxide and suspended particulate reagent-iminodiacetate (SPR-IDA) and Xu et al. (2013) using ZrO-Chelex®100 as a mixed binding gel to remove phosphate from a solution containing various phosphate concentrations. The 0.8 mg/L phosphate solution gave the highest adsorption efficiency ($79.3 \pm 0.7\%$). When a low phosphate concentration is used, binding sites on the surface of the mixed binding gel are available (Jiang et al., 2013). H. Ding et al. (2017) also state that high phosphate adsorption capacity can be achieved when the phosphate concentration is low. The repulsion between phosphate ions and the binding agent with increased phosphate ions in the solution also affects phosphate adsorption on the mixed binding gel.

Elution Efficiencies

Phosphate elution from the ferrihydrite-mixed binding gel is generally done using a 0.25 M H_2SO_4 (Wu, Wang, Jiao, & Wu, 2014). However, when 0.3 M H_2SO_4 solution was used to elute phosphate from the Fe_3O_4 -Chelex®100 mixed binding layer, the elution efficiency was only $3.9 \pm 0.8\%$. Despite using several acids and increasing the acid concentration, the elution efficiency remained below 10%. Since 1 M NaOH is typically used for phosphate elution from Zr-oxide binding gel, NaOH was used as an elution solution (S. Ding et al., 2016).

Table 1 shows that the phosphate elution efficiency of mixed binding gels using acid and base solutions differed. Only a tiny fraction of phosphate was released from the mixed binding gel using acidic solutions. On the contrary, the phosphate elution efficiency reached $\geq 70\%$ when a basic solution was used. This finding showed that alkaline solutions are more effective in facilitating phosphate elution from mixed binding gels.

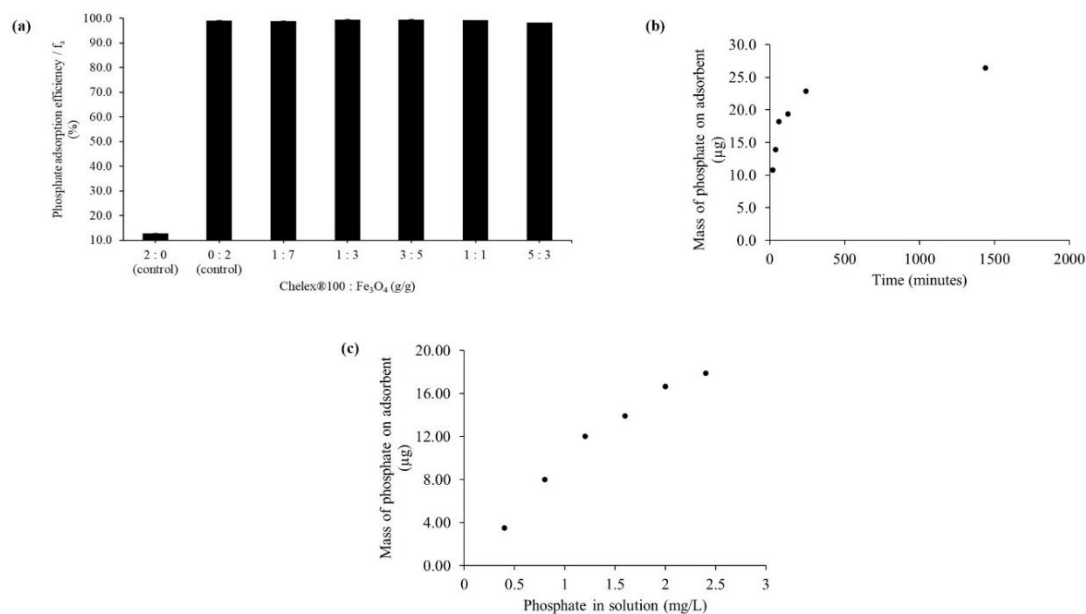


Figure 2. Effect of (a) binding agent composition (Chelex®100: Fe_3O_4), (b) contact time, and (c) initial concentration of phosphate in solution on phosphate adsorption efficiency

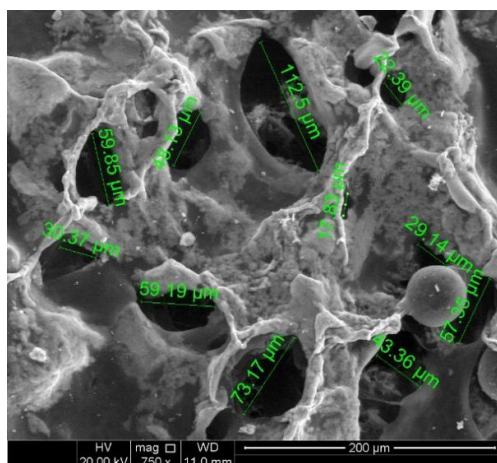


Figure 3. SEM image of Chelex®100: Fe_3O_4 3:5 impregnated in agarose-oxalic acid gel

Table 1 Elution efficiencies (f_e) of phosphate from Fe_3O_4 /Chelex®100-agarose oxalic acid gel in acidic and alkaline solutions

Solution	Concentration (M)	f_a^a (%)	f_e (%)
Acid			
H_2SO_4	0.3	60.5 ± 0.5	3.9 ± 0.8
	1	74.5 ± 1.2	4.3 ± 0.1
HNO_3	1	74.9 ± 1.4	6.0 ± 0.3
HCl	1	70.1 ± 0.8	7.3 ± 0.2
Base			
NaOH	0.1	91.4 ± 0.4	70.0 ± 0.2
	0.2	92.6 ± 1.1	86.6 ± 1.3
	0.3	94.7 ± 0.6	89.1 ± 0.7
	0.4	97.5 ± 0.6	95.3 ± 0.4
	0.5	98.5 ± 0.4	83.7 ± 2.1
	1	94.1 ± 0.6	$95.0^b \pm 0.7$

$^a f_a$ is adsorption efficiencies of 2 mg phosphate/L (deionized water) in Fe_3O_4 -agarose-oxalic acid gels for 24 hours. b the solution became turbid after the reagent for phosphate analysis was added.

Phosphate could be released from the gel by substituting it with OH^- ion from NaOH solution at the binding site (Fe-OH or $-\text{Fe}_2\text{O}$) in Fe_3O_4 (Karunanayake et al., 2019). Low OH^- concentrations were inadequate to release all the phosphate from the binding gel. The higher the NaOH concentration, the more phosphate is released.

However, as NaOH concentration increases, phosphate elution efficiency decreases. Increasing NaOH concentration increases the quantity of OH^- ions in the solution. The increasing number of OH^- ions creates intense competition between phosphate and OH^- ions to occupy adsorption sites on the Fe_3O_4 surface. Therefore, free hydroxyl ions can stimulate and enhance phosphate ion adsorption (Ajmal et al., 2018). Conversely, it is difficult for phosphate ions to desorb from Fe_3O_4 because they form monodentate and bidentate complexes with the positive sites on Fe_3O_4 , whose bonds are stronger than electrostatic interactions.

The most effective eluent for phosphate elution was 0.4 M NaOH solution, having an efficiency of $95.3 \pm 0.4\%$. This result outperforms the phosphate elution efficiency of Chelex®100-Metsorb using 1 M NaOH and Chelex®100-ZrO-agarose gel using 0.2 M NaOH - 0.5 M H_2O_2 (Panther et al., 2014; Wang et al., 2017). **Figure 4a** confirms that increasing elution volume from 15 to 20 mL results in 9% higher f_e values. 20 mL of 0.4 M NaOH solution eluted phosphate from the mixed binding gel by $100.2 \pm 0.6\%$. Phosphate decreased when a higher volume of 0.4 M NaOH solution was used due to the electrostatic repulsion of OH^- ions. As such, 20 mL of 0.4 M NaOH solution was chosen for the next step.

The effect of elution time on the phosphate elution factor of the mixed binding gel is presented in Figure 4b—the longer the gel is in contact with the elution solution, the more phosphate is eluted until the highest ($100.4 \pm 0.3\%$) at 60 minutes. After 60 minutes, Al, Cd, Co, Cr, Cu, Ni, Pb, and Zn were almost entirely eluted (Devillers et al., 2017). There was a fluctuation in phosphate elution (4–6%) at 120 and 240 minutes. Nevertheless, after 24 hours of elution time, phosphate elution returned to $100.7 \pm 0.9\%$.

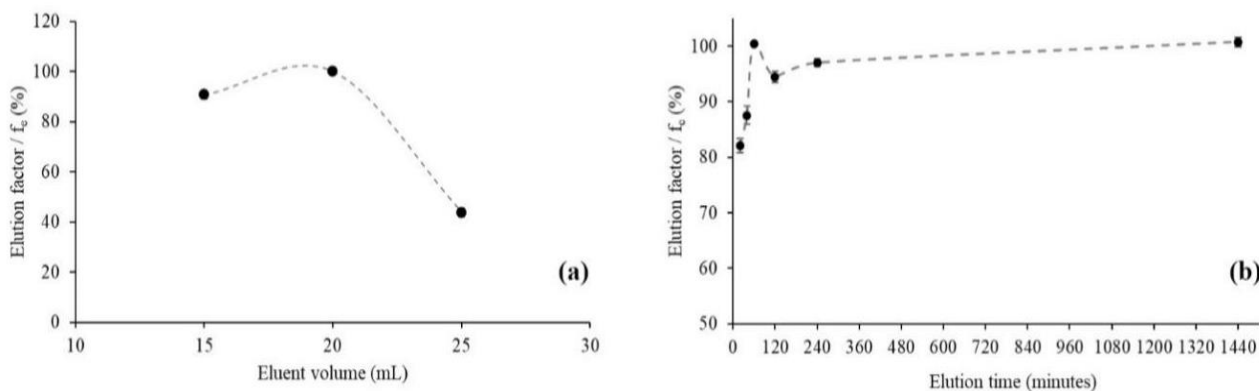


Figure 4. Effect of (a) eluent volume and (b) elution time on phosphate elution efficiency from mixed binding gel

DGT Assembly

The diffusion coefficient was calculated after graphing the relationship between accumulated mass and deployment time (**Figure 5**). The phosphate diffusion coefficient using diffusive gel stored in distilled water combined with the mixed binding gel using the DGT time-series method was $(4.29 \pm 1.08) \times 10^{-6} \text{ cm}^2/\text{s}$. The diffusion coefficient of phosphate was $(6.56 \pm 0.95) \times 10^{-6} \text{ cm}^2/\text{s}$ when the diffusion gel was kept in 0.1 M NaNO_3 solution. This finding showed that the diffusion gel storage solution affected phosphate diffusion. Storing diffusive gels in an electrolyte solution positively impacts phosphate diffusion coefficients. Diffusive gels soaked in electrolyte solutions, such as 0.1 M NaNO_3 , reduce the negative charge on the surface of agarose gel (Wang et al., 2016), allowing phosphate to pass through the gel more easily.

Binding agents and gel types also affect phosphate diffusion coefficients. Zr-oxide combined with A-62 MP and T-42H resin impregnated in agarose produced a phosphate diffusion coefficient of $6.6 \times 10^{-6} \text{ cm}^2/\text{s}$ (M. Ren et al., 2020), while Wang et al. (2016) obtained $(6.86 \pm 0.03) \times 10^{-6} \text{ cm}^2/\text{s}$ using ZrO-Chelex®100-agarose gel. These values are slightly higher than the phosphate diffusion coefficient in APA-gel DGT as the standard gel ($6.05 \times 10^{-6} \text{ cm}^2/\text{s}$) due to the agarose pore size ($>20 \text{ nm}$) being larger than APA gel ($>5 \text{ nm}$).

Table 2 compares the phosphate diffusion coefficients in water using the DGT technique. Diffusive layers employed in the table include gel and dialysis membrane. Table 2 shows that the phosphate diffusion coefficient through the gel is higher than the dialysis membrane because the pore size of the dialysis membrane is smaller than the pore size of the gel (W. Li et al., 2014). As previously stated, the binding agent affected the phosphate diffusion coefficient. The Fe_3O_4 /Chelex®100 binding agent used in this study gave the highest phosphate diffusion coefficient and its value closest to the APA gel compared to previous studies. The phosphate diffusion coefficient close to the APA gel can accurately measure phosphate concentration using the DGT technique.

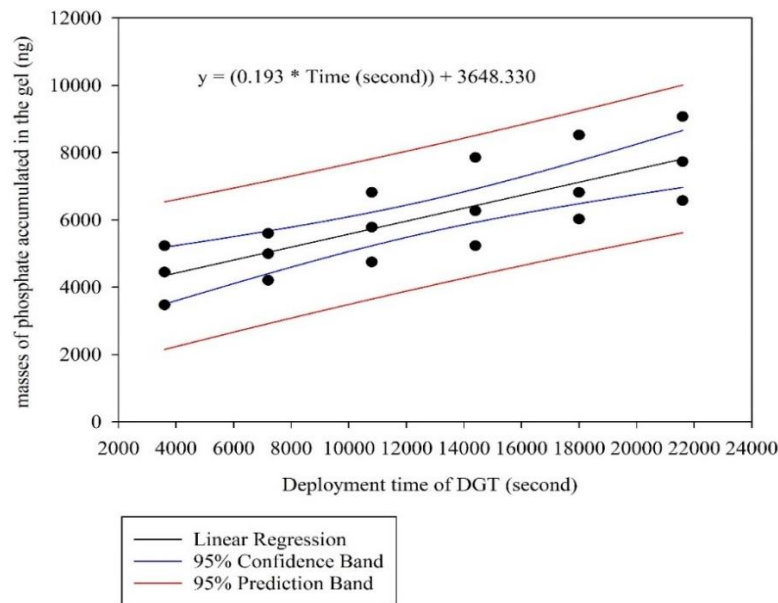


Figure 5. Average phosphate diffusion coefficient determination

Table 2. Comparison of phosphate diffusion coefficients in water using DGT

Diffusive Layer	Binding Layer	Method	Diffusion Coefficient (cm^2/s)	Reference
Agarose-oxalic acid gel	Chelex®100- Fe_3O_4 in agarose-oxalic acid gel	DGT Time Series	6.56×10^{-6}	This study
APA gel	Ferrihydrite gel	Diffusion Cell, DGT Time Series	6.05×10^{-6}	H. Zhang et al. (1998)
Agarose gel	Metsorb in agarose gel	DGT Time Series	5.46×10^{-6}	J. Huang et al. (2017)
Bis-poliakrilamid (BPA) gel	La-MOF in BPA gel	DGT Time Series	2.216×10^{-6}	Santikasari et al. (2020)
Dialysis membrane	Polyquaternary ammonium salt (PQAS) aqueous solution	Diffusion Cell	1.95×10^{-6}	Chen et al. (2014)
Dialysis membrane	Polydiallyldimethylammonium chloride (PDA) aqueous solution	Diffusion Cell	2.45×10^{-6}	W. Li et al. (2014)
Dialysis membrane	Zr-based metal-organic frameworks liquid	Diffusion Cell	1.55×10^{-7}	Qin et al. (2018)
Dialysis membrane	Fe_3O_4 NPs suspension	Diffusion Cell, DGT Time Series	9.5×10^{-8} (DGT Time Series) 1.04×10^{-7} (Diffusion Cell)	Y. Zhang et al. (2018)

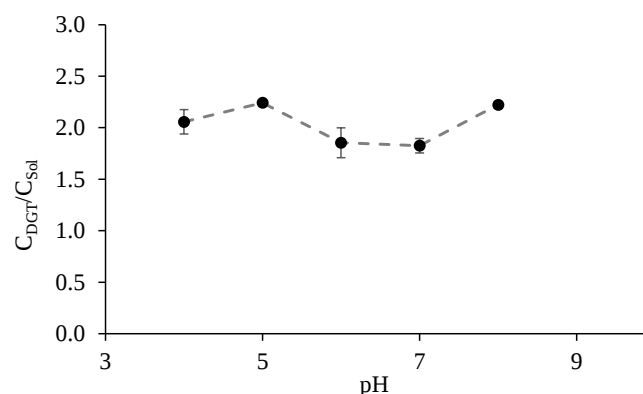


Figure 6. Values of C_{DGT}/C_{sol} for phosphate under different pH conditions

Table 3. Values of C_{DGT}/C_{sol} for phosphate under different NaNO_3 concentrations

NaNO_3 (M)	C_{DGT}/C_{sol}
0.01	1.89 ± 0.01
0.1	1.85 ± 0.04

DGT Performance

The blank Fe_3O_4 -Chelex®100-DGTma at pH 4 – 8 ranged from $0.162 - 1.238 \mu\text{g}$ with MDL of 0.03 mg/L. The mixed binding gel blanks were much higher than Biochar-Zeolite-ZrO gel ($\sim 220 \text{ ng}$ for P) (Feng et al., 2018), indicating that the phosphate content in the mixed binding gel cannot be ignored. Contamination can come from reagents used for binding agent synthesis or during the washing and drying, as occurred with ZrO_2 (Eismann et al., 2023). The blank indicated phosphate contamination for 0.01 – 0.1 M NaNO_3 ($0.913 - 0.933 \mu\text{g}$) with an MDL of 0.03 mg/L. Nevertheless, the detection limit in this study was lower than the phosphate threshold of 1 mg/L as established by the Republic of Indonesia Government Regulation Number 22 of 2021, demonstrating that phosphate in water can be monitored using the Fe_3O_4 -Chelex®100-DGT method.

Figure 6 shows the effect of solution pH on C_{DGT}/C_{sol} . The phosphate measurements of mixed binding gel-DGT in pH 4 - 8 solution were approximately twice from the grab sampling phosphate concentration. Similar results were obtained for phosphate measurement in 0.01 and 0.1 M NaNO_3 (Table 3). C_{DGT}/C_{sol} of ~ 2.2 at pH 3 was obtained using Fe_3O_4 NP liquid binding agent. The phosphate accumulation in mixed binding gel-DGT decreased as the pH of the solutions increased until C_{DGT}/C_{sol} remained constant at pH > 7. The increase in C_{DGT}/C_{sol} when studying the effect of ionic strength was not only caused by the strength of the binding agent to adsorb phosphate but also due to the salt effect, which influenced the ionic activity coefficient of phosphate (Y. Zhang et al., 2018).

The performance of mixed binding layer DGT was validated in natural freshwater. The equation $y = 274.21x$ ($R^2 = 0.944$) was obtained from the calibration curve. Based on the equation, the phosphate concentration in natural freshwater was $0.505 \pm 0.097 \text{ mg/L}$, whereas the grab sampling method yielded a phosphate concentration of $0.454 \pm 0 \text{ mg/L}$. A statistical test compared the DGT-measured phosphate concentration to the phosphate concentration acquired via grab sampling. Statistical analysis at a 95% confidence level using the T-test revealed $t_{\text{count}} (1.04) < t_{\text{table}} (4.03)$. This result indicated that the newly developed mixed binding gel-DGT is not significantly different from the grab sampling method, globally recognized as a standard method.

CONCLUSIONS

Chelex®100 and Fe_3O_4 impregnated in one binding layer enable several analytes to be measured with a single DGT passive sampler. The important finding of this study is that Fe_3O_4 /Chelex®100-Agarose-Oxalic Acid hydrogel as a binding gel in a DGT sampler can effectively adsorb phosphate, and the diffusion coefficient of phosphate using this gel is comparable to the earlier studies. The highest phosphate adsorption efficiency was given by the composition of Chelex®100: Fe_3O_4 3:5, phosphate concentration 0.8 mg/L, and 4-hour adsorption time ($79.3 \pm 0.7\%$). SEM image of mixed binding gel showed pore sizes of $22.39 - 112.5 \mu\text{m}$ with heterogeneous distribution. High phosphate elution efficiency was obtained using 20 mL of 0.4 M NaOH solution with a 1-hour elution time ($100.4 \pm 0.3\%$). The phosphate diffusion coefficient by Fe_3O_4 /Chelex®100-agarose-oxalic acid-DGT was $6.56 \times 10^{-6} \text{ cm}^2/\text{s}$. The phosphate concentration measurements using mixed binding gel-DGT in solution at pH 4-8 and NaNO_3 0.01 – 0.1 M was approximately twice the directly measured phosphate concentration. Deployment in natural water showed that Fe_3O_4 /Chelex®100-agarose-oxalic acid-DGT is a very good passive sampler for phosphate measurement.

ACKNOWLEDGEMENTS

The authors thank the Ministry of Education, Culture, Research, and Technology for funding this research through the Doctoral Dissertation Research Grant 2022-2023.

REFERENCES

Ajmal, Z., Muhmood, A., Usman, M., Kizito, S., Lu, J., Dong, R., & Wu, S. (2018). Phosphate removal from aqueous solution using iron oxides: Adsorption, desorption and regeneration characteristics. *Journal of Colloid and Interface Science*, 528, 145–155. <https://doi.org/10.1016/j.jcis.2018.05.084>

Bajagain, R., Noh, S., Kim, Y., Kim, H., Seok, K., Bailon, M. X., & Hong, Y. (2024). Application of diffusive gradient in thin film probes to monitor trace levels of labile methylmercury in freshwaters. *Environmental Monitoring and Assessment*, 196(4), 404. <https://doi.org/10.1007/s10661-024-12564-8>

Booij, K., Robinson, C. D., Burgess, R. M., Mayer, P., Roberts, C. A., Ahrens, L., Whitehouse, P.

- (2016). Passive sampling in regulatory chemical monitoring of nonpolar organic compounds in the aquatic environment. *Environmental Science & Technology*, 50(1), 3–17. <https://doi.org/10.1021/acs.est.5b04050>
- Cao, H., Bu, Q., Li, Q., Yang, L., Tang, J., & Yu, G. (2024). Evaluation of the DGT passive samplers for integrating fluctuating concentrations of pharmaceuticals in surface water. *Science of The Total Environment*, 926, 172067. <https://doi.org/10.1016/j.scitotenv.2024.172067>
- Chen, H., Zhang, M.-H., Gu, J.-L., Zhao, G., Zhang, Y., & Li, J.-R. (2014). Measurement of dissolved reactive phosphorus in water with polyquaternary ammonium salt as a binding agent in diffusive gradients in thin-films technique. *Journal of Agricultural and Food Chemistry*, 62(50), 12112–12117. <https://doi.org/10.1021/jf5040702>
- De Rezende Bonesio, M., & Pissetti, F. L. (2020). Magnetite particles covered by amino-functionalized poly(dimethylsiloxane) network for copper(II) adsorption from aqueous solution. *Journal of Sol-Gel Science and Technology*, 94(1), 154–164. <https://doi.org/10.1007/s10971-019-05154-5>
- Ding, H., Zhao, Y., Duan, Q., Wang, J., Zhang, K., Ding, G., Ding, C. (2017). Efficient removal of phosphate from aqueous solution using novel magnetic nanocomposites with Fe₃O₄@SiO₂ core and mesoporous CeO₂ shell. *Journal of Rare Earths*, 35(10), 984–994. [https://doi.org/10.1016/S1002-0721\(17\)61003-2](https://doi.org/10.1016/S1002-0721(17)61003-2)
- Ding, S., Xu, D., Wang, Y., Wang, Y., Li, Y., Gong, M., & Zhang, C. (2016). Simultaneous measurements of eight oxyanions using high-capacity diffusive gradients in thin films (Zr-oxide DGT) with a high-efficiency elution procedure. *Environmental Science & Technology*, 50(14), 7572–7580. <https://doi.org/10.1021/acs.est.6b00206>
- Eismann, C. E., Menegário, A. A., Geraldo De Oliveira, E., Elias, L. P., Martins Viana, J. L., Teixeira Zanatta, M. B., Fostier, A. H. (2023). A simple, green and low-cost agarose-based binding layer for simultaneous determination of cations and anions in aquatic systems using DGT. *Analytical Methods*, 15(27), 3310–3317. <https://doi.org/10.1039/D3AY00682D>
- Fadli, A., Komalasari, Adnan, A., Iwantono, Rahimah, & Addabsi, A. S. (2019). Synthesis of magnetite nanoparticles via Co-precipitation method. *IOP Conference Series: Materials Science and Engineering*, 622(1), 012013. <https://doi.org/10.1088/1757-899X/622/1/012013>
- Feng, Z., Wang, N., He, M., Yang, L., Wang, Y., & Sun, T. (2018). Simultaneous sampling of dissolved orthophosphate and ammonium in freshwaters using diffusive gradients in thin films with a mixed binding phase. *Talanta*, 186, 176–182. <https://doi.org/10.1016/j.talanta.2018.04.045>
- Franco, D., Lee, J., Arbelaez, S., Cohen, N., & Kim, J.-Y. (2017). Removal of phosphate from surface and wastewater via electrocoagulation. *Ecological Engineering*, 108, 589–596. <https://doi.org/10.1016/j.ecoleng.2017.07.031>
- Hou, L., Liang, Q., & Wang, F. (2020). Mechanisms that control the adsorption–desorption behavior of phosphate on magnetite nanoparticles: The role of particle size and surface chemistry characteristics. *RSC Advances*, 10(4), 2378–2388. <https://doi.org/10.1039/C9RA08517C>
- Huang, J., Bennett, W. W., Teasdale, P. R., Kankanamge, N. R., & Welsh, D. T. (2017). A modified DGT technique for the simultaneous measurement of dissolved inorganic nitrogen and phosphorus in freshwaters. *Analytica Chimica Acta*, 988, 17–26. <https://doi.org/10.1016/j.aca.2017.08.024>
- Huang, Y., Lee, X., Grattieri, M., Yuan, M., Cai, R., Macazo, F. C., & Minter, S. D. (2020). Modified biochar for phosphate adsorption in environmentally relevant conditions. *Chemical Engineering Journal*, 380, 122375. <https://doi.org/10.1016/j.cej.2019.122375>
- Jiang, H., Chen, P., Luo, S., Tu, X., Cao, Q., & Shu, M. (2013). Synthesis of novel nanocomposite Fe₃O₄/ZrO₂/chitosan and its application for removal of nitrate and phosphate. *Applied Surface Science*, 284, 942–949. <https://doi.org/10.1016/j.apsusc.2013.04.013>
- Karunanayake, A. G., Navarathna, C. M., Gunatilake, S. R., Crowley, M., Anderson, R., Mohan, D., Mlsna, T. (2019). Fe₃O₄ Nanoparticles dispersed on douglas fir biochar for phosphate sorption. *ACS Applied Nano Materials*, 2(6), 3467–3479. <https://doi.org/10.1021/acsanm.9b00430>
- Kreuzeder, A., Santner, J., Prohaska, T., & Wenzel, W. W. (2013). Gel for simultaneous chemical imaging of anionic and cationic solutes using diffusive gradients in thin films. *Analytical Chemistry*, 85(24), 12028–12036. <https://doi.org/10.1021/ac403050f>
- Lenders, J. J. M., Bawazer, L. A., Green, D. C., Zope, H. R., Bomans, P. H. H., De With, G., ... Sommerdijk, N. A. J. M. (2017). Combinatorial evolution of biomimetic magnetite nanoparticles. *Advanced Functional Materials*, 27(10), 1604863. <https://doi.org/10.1002/adfm.201604863>
- Li, C., Ding, S., Yang, L., Wang, Y., Ren, M., Chen, M., ... Lichtfouse, E. (2019). Diffusive gradients in thin films: Devices, materials and

- applications. *Environmental Chemistry Letters*, 17(2), 801–831. <https://doi.org/10.1007/s10311-018-00839-9>
- Li, W., Lee, L. Y., Yung, L. Y. L., He, Y., & Ong, C. N. (2014). Combination of in situ preconcentration and on-site analysis for phosphate monitoring in fresh waters. *Analytical Chemistry*, 86(15), 7658–7665. <https://doi.org/10.1021/ac5015386>
- Lin, B., & Pan, F. (2023). Applications of DGT in coastal sediments: Monitoring and biogeochemical study of trace metals and oxyanions. *Trends in Environmental Analytical Chemistry*, 39, e00207. <https://doi.org/10.1016/j.teac.2023.e00207>
- Marrugo-Madrid, S., Turull, M., Zhang, H., & Díez, S. (2021). Diffusive gradients in thin films for the measurement of labile metal species in water and soils: A review. *Environmental Chemistry Letters*, 19(5), 3761–3788. <https://doi.org/10.1007/s10311-021-01246-3>
- Panther, J. G., Bennett, W. W., Welsh, D. T., & Teasdale, P. R. (2014). Simultaneous measurement of trace metal and oxyanion concentrations in water using diffusive gradients in thin films with a chelex–metsorb mixed binding layer. *Analytical Chemistry*, 86(1), 427–434. <https://doi.org/10.1021/ac402247j>
- Pouran, H. M., Martin, F. L., & Zhang, H. (2014). Measurement of ZnO nanoparticles using diffusive gradients in thin films: Binding and diffusional characteristics. *Analytical Chemistry*, 86(12), 5906–5913. <https://doi.org/10.1021/ac500730s>
- Pouran, H., Perez Colodrero, R., Wu, S., Hix, G., Zakharova, J., & Zhang, H. (2020). Assessment of ATR-FTIR spectroscopy with multivariate analysis to investigate the binding mechanisms of Ag and TiO₂ nanoparticles to Chelex®-100 or Metsorb™ for the DGT technique. *Analytical Methods*, 12(7), 959–969. <https://doi.org/10.1039/C9AY02458A>
- Qin, W., Gu, Y., Wang, G., Wu, T., Zhang, H., Tang, X., Zhao, H. (2018). Zirconium metal organic frameworks-based DGT technique for in situ measurement of dissolved reactive phosphorus in waters. *Water Research*, 147, 223–232. <https://doi.org/10.1016/j.watres.2018.10.003>
- Ren, M., Ding, S., Shi, D., Zhong, Z., Cao, J., Yang, L., ... Wang, Y. (2020). A new DGT technique comprised in a hybrid sensor for the simultaneous measurement of ammonium, nitrate, phosphorus and dissolved oxygen. *Science of The Total Environment*, 725, 138447. <https://doi.org/10.1016/j.scitotenv.2020.138447>
- Ren, S., Li, L., Li, Y., Wu, J., & Dou, Y. (2024). Development and field application of a diffusive gradients in thin-films passive sampler for monitoring three polycyclic aromatic hydrocarbon derivatives and one polycyclic aromatic hydrocarbon in waters. *Water*, 16(5), 684. <https://doi.org/10.3390/w16050684>
- Santikasari, C., Saefumillah, A., & Zulys, A. (2020). Lanthanum based metal organic frameworks as a new binding agent material for diffusive gradient in thin film (DGT) technique in bioavailability of phosphate removal. *IOP Conference Series: Materials Science and Engineering*, 902(1), 012043. <https://doi.org/10.1088/1757-899X/902/1/012043>
- Shakallis, A. G., Fallowfield, H., Ross, K. E., & Whaley, H. (2022). The application of passive sampling devices in wastewater surveillance. *Water*, 14(21), 3478. <https://doi.org/10.3390/w14213478>
- Shiva, A. H., Teasdale, P. R., Bennett, W. W., & Welsh, D. T. (2015). A systematic determination of diffusion coefficients of trace elements in open and restricted diffusive layers used by the diffusive gradients in a thin film technique. *Analytica Chimica Acta*, 888, 146–154. <https://doi.org/10.1016/j.aca.2015.07.027>
- Smolíková, V., Pelcová, P., Ridošková, A., & Leermakers, M. (2022). Diffusive Gradients in Thin-films technique for uranium monitoring along a salinity gradient: A comparative study on the performance of Chelex-100, Dow-PIWBA, Diphonix, and Lewatit FO 36 resin gels in the Scheldt estuary. *Talanta*, 240, 123168. <https://doi.org/10.1016/j.talanta.2021.123168>
- Sun, Q., Zhang, L., Ding, S., Li, C., Yang, J., Chen, J., & Wang, P. (2015). Evaluation of the diffusive gradients in thin films technique using a mixed binding gel for measuring iron, phosphorus and arsenic in the environment. *Environmental Science: Processes & Impacts*, 17(3), 570–577. <https://doi.org/10.1039/C4EM00629A>
- Taylor, A. C., Fones, G. R., & Mills, G. A. (2020). Trends in the use of passive sampling for monitoring polar pesticides in water. *Trends in Environmental Analytical Chemistry*, 27, e00096. <https://doi.org/10.1016/j.teac.2020.e00096>
- Wang, Y., Ding, S., Gong, M., Xu, S., Xu, W., & Zhang, C. (2016). Diffusion characteristics of agarose hydrogel used in diffusive gradients in thin films for measurements of cations and anions. *Analytica Chimica Acta*, 945, 47–56. <https://doi.org/10.1016/j.aca.2016.10.003>
- Wang, Y., Ding, S., Ren, M., Li, C., Xu, S., Sun, Q., & Xu, L. (2019). Enhanced DGT capability for measurements of multiple types of analytes using synergistic effects among different

- binding agents. *Science of The Total Environment*, 657, 446–456. <https://doi.org/10.1016/j.scitotenv.2018.12.016>
- Wang, Y., Ding, S., Shi, L., Gong, M., Xu, S., & Zhang, C. (2017). Simultaneous measurements of cations and anions using diffusive gradients in thin films with a ZrO-Chelex mixed binding layer. *Analytica Chimica Acta*, 972, 1–11. <https://doi.org/10.1016/j.aca.2017.04.007>
- Wu, Z., Wang, S., Jiao, L., & Wu, F. (2014). The simultaneous measurement of phosphorus, sulfide, and trace metals by ferrihydrite/AgI/Chelex-100 DGT (diffusive gradients in thin films) probe at sediment/water interface (SWI) and remobilization assessment. *Water, Air, & Soil Pollution*, 225(11), 2188. <https://doi.org/10.1007/s11270-014-2188-2>
- Wulandari, I. O., Mardila, V. T., Santjojo, D. J. D. H., & Sabarudin, A. (2018). Preparation and characterization of chitosan-coated Fe₃O₄ nanoparticles using ex-situ co-precipitation method and tripolyphosphate/sulphate as dual crosslinkers. *IOP Conference Series: Materials Science and Engineering*, 299, 012064. <https://doi.org/10.1088/1757-899X/299/1/012064>
- Wurtsbaugh, W. A., Paerl, H. W., & Dodds, W. K. (2019). Nutrients, eutrophication and harmful algal blooms along the freshwater to marine continuum. *WIREs Water*, 6(5), e1373. <https://doi.org/10.1002/wat2.1373>
- Xu, D., Chen, Y., Ding, S., Sun, Q., Wang, Y., & Zhang, C. (2013). Diffusive gradients in thin films technique equipped with a mixed binding gel for simultaneous measurements of dissolved reactive phosphorus and dissolved iron. *Environmental Science & Technology*, 130827135544005. <https://doi.org/10.1021/es401822x>
- Zhang, C., Ding, S., Xu, D., Tang, Y., & Wong, M. H. (2014). Bioavailability assessment of phosphorus and metals in soils and sediments: A review of diffusive gradients in thin films (DGT). *Environmental Monitoring and Assessment*, 186(11), 7367–7378. <https://doi.org/10.1007/s10661-014-3933-0>
- Zhang, H., & Davison, W. (1999). Diffusional characteristics of hydrogels used in DGT and DET techniques. *Analytica Chimica Acta*, 398(2–3), 329–340. [https://doi.org/10.1016/S0003-2670\(99\)00458-4](https://doi.org/10.1016/S0003-2670(99)00458-4)
- Zhang, H., Davison, W., Gadi, R., & Kobayashi, T. (1998). In situ measurement of dissolved phosphorus in natural waters using DGT. *Analytica Chimica Acta*, 370(1), 29–38. [https://doi.org/10.1016/S0003-2670\(98\)00250-5](https://doi.org/10.1016/S0003-2670(98)00250-5)
- Zhang, Y., Song, J., Zhou, H., Zhang, Y., & Wang, G. (2018). Novel Fe₃O₄ nanoparticles-based DGT device for dissolved reactive phosphate measurement. *New Journal of Chemistry*, 42(4), 2874–2881. <https://doi.org/10.1039/C7NJ04464J>