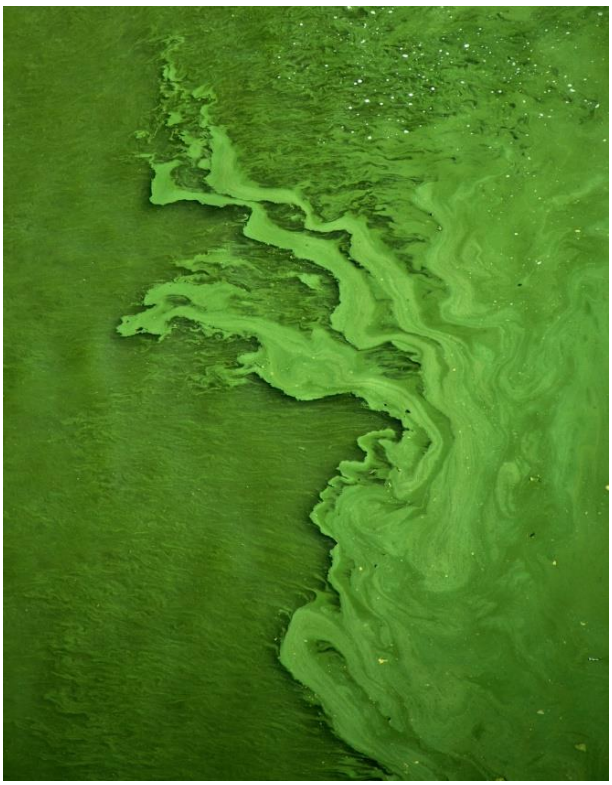
# Phosphorus information for Wye catchment citizen scientists

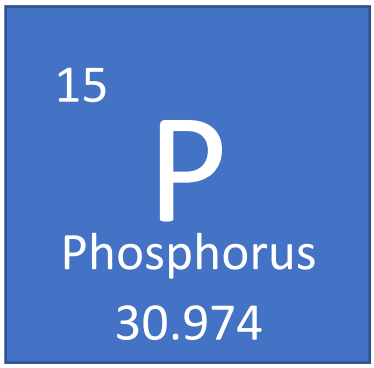
## What is phosphorus and why is nutrient pollution an issue for rivers?

**Phosphorus** (P) is an element in the periodic table and is an essential nutrient for life. P naturally exists in the environment because it is stored in rock and is made available for biological uptake through the process of weathering or by volcanic activity. In freshwater bodies, P is often the key 'biolimiting' nutrient which means it is the nutrient whose availability determines the rate of plant and algal growth.

P is found in high concentrations in some agricultural run-off and in wastewater. In environments where P is the biolimiting nutrient, if the amount of P entering a river increases (and other conditions are right) it is likely to lead to the dramatic growth of algae, resulting in algal blooms and an undesirable disturbance to the balance of organisms present in the water. This process, known as eutrophication, is shown in Image 2. Certain algal blooms also produce toxins (known as toxic algal blooms) which may be harmful to other organisms, such as humans, livestock and pets, who come in contact with the waterbody. Sometimes Nitrogen (N) is the biolimiting nutrient rather than P in freshwater systems, but this is much less common.



Algal bloom
Photo Credit: Mihály Köles
*Vlatava with massive algae growth on Unsplash*



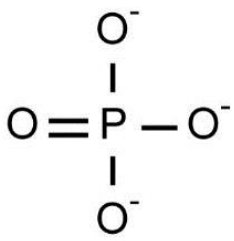
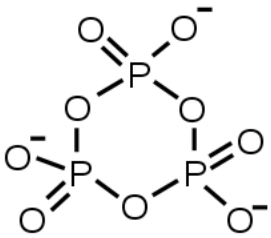
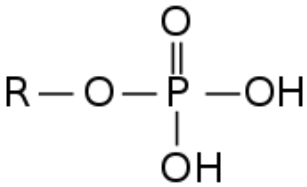

Dead fish in algal bloom
Photo Credit: Dr. Jennifer L. Graham | U.S. Geological Survey
*eutrophication&hypoxia on Flickr*

## What different chemical forms of phosphorus exist in water?

In water, phosphorus always occurs as a compound combined with oxygen, and sometimes also with other elements, but never in its elemental form (P). When phosphorus combines with oxygen it forms phosphate. Many different types of phosphate exist but they can be categorised into three main groups:

- Orthophosphate
- Condensed phosphate
- Organic phosphate

**Table 1.** Types of phosphate that exist in water

| | **Orthophosphate** | **Condensed phosphate** | **Organic phosphate** |
|---|---|---|---|
| **Type of compound** | Free phosphate ion ($PO_4^{3-}$) | Multiple orthophosphate molecules combined together, sometimes also with other inorganic molecules such as salts, minerals or metals | Phosphate contained within or bound to an organic[1] compound |
| **Example** |  Orthophosphate ion [1] |  Metaphosphate [1] |  Organic phosphate [2] (where R is an organic molecule) |
| **Chemistry** | Inorganic | Inorganic | Organic |
| **State in water** | Dissolved | Dissolved or suspended | Dissolved or suspended |
| **Bioavailability[3]** | Bioavailable | Partially bioavailable | Partially bioavailable |

[1] Substance containing C-H bonds; [2] Image credit: Hach;
[3] Degree to which a substance can be taken up by an organism

## What different forms of phosphorus can be measured in water?

Measuring the amount of phosphorus in water can be difficult as no perfect methods exist to measure the main chemical forms of phosphate (described above in Table 1). Instead, three main terms are used to describe the fraction of P measured using the most common methods of analysis:

- Reactive phosphorus (or soluble reactive phosphorus if filtered)
- Acid hydrolysable phosphorus
- Total phosphorus

## Reactive Phosphorus

- Reactive Phosphorus (RP) is a term used to describe what is actually measured when we attempt to measure **orthophosphate ($PO_4^{3-}$)** in a water sample.
- This method involves adding molybdate to the sample which reacts with the orthophosphate present. A coloured compound forms in solution and the intensity of the colour in the reacted sample indicates the concentration of orthophosphate present. The reaction is carried out under acidic conditions by adding ascorbic acid to reduce oxidation of the compound that forms.
- Small amounts of condensed phosphates (which also react with the reagent) may be measured in addition to free orthophosphate when using this method.

## Acid Hydrolysable Phosphorus

- Acid Hydrolysable Phosphorus is a term used to describe what is actually measured when we attempt to measure the amount of **orthophosphate and condensed phosphate** in a water sample.
- This method involves adding sulphuric acid to a sample and then heating it to 'digest' (break the bonds between orthophosphate and the salts it is bound to) the condensed phosphate in the sample and convert it into orthophosphate. The total amount of orthophosphate (**$PO_4^{3-}$**) present in the sample is then measured using the molybdate method, explained above.
- Small amounts of organic phosphates (which are also hydrolyzed by the acid) are likely to be measured in addition to orthophosphate and condensed phosphate when using this method.

## Total Phosphorus

- Total phosphorus (TP) is a term used to describe what is measured when we attempt to measure **all three type of phosphate: orthophosphate, condensed phosphate and organic phosphate**, present in a water sample.
- This method involves converting condensed phosphate and organic phosphate into orthophosphate, by digesting the sample with heat and acid, and then adding a strong oxidizing agent. The heat, acid and oxidizing agent break the strong bonds in the organic molecule. The total amount of orthophosphate (**$PO_4^{3-}$**) present in the sample is then measured using the molybdate method, explained above.
- This method gives us an idea of the total amount of phosphate (and therefore phosphorus) present in a sample but, like the other methods, is imperfect and won't capture all P.

## What methods can be used by citizen scientists to monitor phosphorus?

All test kits used by citizen scientists to assess phosphorus concentrations in the field will be measuring **reactive phosphorus**. This is because to assess acid hydrolysable phosphorus and total phosphorus, it is necessary to heat samples and add strong oxidizing agents. This is difficult to do and only possible using specialist laboratory equipment and potentially hazardous chemical processing. Reactive phosphorus, as an indicator of orthophosphate, is the simplest fraction to measure and arguably also the most useful since it represents the most bioavailable form of phosphorus found in water.

Three test kits used by citizen scientists across the Wye catchment to assess phosphorus concentrations are: La Motte phosphate test strips, Hanna phosphate low-range checker, and Hach Pocket Colorimeter. Details of these test kits are set out in Table 2, including the manufacturers' reported range, resolution and accuracy. We are working with citizen science and professional data to better understand the difference in the quality of data obtained from the different test kits used. **It is critical that Citizen Science Coordinators for each group refer to the manufacture's hazard warnings for each test, understand the risks of the reagents used and take full account of them when writing risk assessments and advising citizen scientists of the precautions that must be taken during field testing.**

## How is the concentration of phosphorus in water reported?

Phosphorus concentrations in water are recorded in units of ppm(mg/L) or ppb(µg /l). These units can easily be converted:
1ppm = 1mg/l ; 1ppb = 1 µg/l; 1ppm= 1000ppb

Phosphorus concentrations are reported either in terms of $PO_4^{3-}$ ("orthophosphate") or in terms of $PO_4^{3-}$-P ("orthophosphate as phosphorus"). The difference is that $PO_4^{3-}$ takes account of both the phosphorus (P) and the oxygen (O) present in the compound, whilst $PO_4^{3-}$-P only considers the phosphorus (P) present in the compound.

The agencies usually report concentrations in terms of $PO_4^{3-}$-P ("orthophosphate as phosphorus"), and this is also what phosphorus standards are most commonly set as. Conversely, most citizen science test kits report results as $PO_4^{3-}$ ("orthophosphate"). These units can be easily converted because a molecule of orthophosphate ($PO_4^{3-}$) weighs 3.06 times more than a molecule of just phosphorus (P). Dividing your "orthophosphate" result by 3.06 can calculate P.

**However, the most useful thing citizen scientists can do is to record results in the exact form that they are reported in by the test kit being used. Don't try to convert between units (ppm to ppb) or from "orthophosphate" to "orthophosphate as P" as this is likely to introduce errors to the data sets.** Any necessary conversions can be done by data user(s) during data analysis.

**Table 2.** Citizen science test kits used for assessing phosphate in river water samples used by groups across the Wye catchment

| Test kit | Form of P measured | Guidance for use | Method | Hazard warnings | Reports P as | Range[1] | Resolution[2] | Accuracy[3] | Cost |
|---|---|---|---|---|---|---|---|---|---|
| La Motte test strips (Phosphate) | Reactive phosphorus | La Motte: https://lamotte.com/amfile/file/download/file/641/product/37/ Cardiff (pg.29): https://drive.google.com/file/d/1kVcZbfqN_yoRLu9lpYwEPaf9VitzSmQ5/view?usp=sharing | Not specified<br><br>No additional reagent required | None provided by manufacturers | **$PO_4^{3-}$** "Ortho-phosphate"<br><br>**ppb** "parts per billion" | 0 - 2500 ppb | 0, 100, 200, 300, 500, 1000, and 2500 ppb | Not stated | Low (~£0.30 per test) |
| Hanna Phosphate Low-range Checker (HI-713) | Reactive phosphorus | Hanna: https://www.hannainstruments.co.uk/pocket-checker-for-phosphate-testing.html#manuals Cardiff (pg. 30-33): https://drive.google.com/file/d/1kVcZbfqN_yoRLu9lpYwEPaf9VitzSmQ5/view?usp=sharing | Adaptation of Standard Method 4500-P E.[4]<br><br>**Reagent required:** HI-713-25 | **See safety data sheet for reagents:** https://www.hannainstruments.co.uk/amfilerating/file/download/file_id/2630/ | **$PO_4^{3-}$** "Ortho-phosphate"<br><br>**ppm** "parts per million" | 0.00 - 2.50 ppm | 0.01 ppm | +/-0.04ppm / ±4% of reading @ 25°C (as stated by manufacturer) | Medium (~£70 + reagents) |
| Hach pocket colorimeter-Phosphate (DR300) | Reactive phosphorus | Hach: https://uk.hach.com/asset-get.download.jsa?id=55721669801 Cardiff (video): https://drive.google.com/file/d/1qkH561tAdviLpRqjYKgZOh2jfrbv8mH0/view?usp=sharing | Adaptation of Standard Method 4500-P E.[4]<br><br>**Reagent required:** Hach PhosVer 3 Powder Pillows | **See safety data sheet for reagents:** https://www.accomn.com/userfiles/modules/file_upload_library_3/1726/PhosVer_3_Phosphate_Reagent.pdf | **$PO_4^{3-}$** "Ortho-phosphate"<br><br>**mg/L** "milligrams per litre" | 0.02 - 3.00 mg/L | 0.01 mg/L | Not stated | High (~£590 + reagents) |

[1] lowest and highest values that can be detected; [2] the smallest change in concentration that can be detected; [3] the closeness of a measurement to the true value; [4] an overview of this standard method can be found online at https://www.nemi.gov/methods/method_summary/7436/

## How should samples be prepared for phosphorus analysis?

### Filtering

Phosphates in water can exist either dissolved or suspended in solution. To separate these two mechanical fractions, a 0.45 micron filter is commonly used. However, this process is not perfect and filtering will still allow some very small suspended (colloidal) phosphates through.

All orthophosphate exists dissolved in solution. When measuring orthophosphate, if the sample is not filtered some particulate P could also be measured. If the sample is filtered before analysis then it is known as soluble reactive phosphorus (SRP), rather than just reactive phosphorus (RP) or total reactive phosphorus (TRP). **Most test kits (including those listed in Table 2) do not require you to filter samples in preparation. It is important to state on your survey if a sample has (for any reason) been filtered before analysis because it may affect the result.**

When acid hydrolysable phosphate or total phosphorus (TP) is assessed the sample is not typically filtered as the process of filtering removes some of the suspended condensed and/or suspended organic fractions. If these samples are filtered before analysis then they are known as *soluble* acid hydrolysable phosphate or total *soluble* phosphorus, rather than just acid hydrolysable phosphate or total phosphorus.

### Settling

Leaving samples to 'settle out' (if they are turbid) or to stand before analysis (if you are busy) can be tempting. However, there are a number of errors that can occur if the sample is left to stand for too long before analysis. Phosphate in the sample will begin to react with other molecules (changing form) or settle out.

**You should analyse samples as soon as possible after collection. It is important to state clearly on your survey if (for any reason) you have left the sample for more than one hour before analysis.**

## What other methods are being used to assess phosphorus concentrations?

Other organisations working across the Wye, including Natural Resources Wales (NRW), the Environment Agency (EA), Dŵr Cymru Welsh Water (DCWW) and the Wye and Usk Foundation (WUF) are monitoring phosphorus concentrations in rivers and streams. These organisations have the capacity to measure total phosphorus as well as reactive phosphorus through carrying out digestion of condensed and organic phosphates. Table 3 shows a summary of the various methods used by organisations across the Wye catchment to measure both reactive and total phosphorus in the laboratory or field environment.

## What are the standards for phosphorus in rivers?

Different rivers, or reaches of a river, across a catchment may have different phosphorus targets set because natural concentrations in the environment are expected to vary between waterbodies, and different ecosystems are able to cope with different amounts of phosphorus. Targets are also set in different ways for SAC and non-SAC rivers. Phosphorus standards for freshwater bodies are typically reported as $PO_4$-P "Orthophosphate as P".

Phosphorus targets for English and Welsh SAC rivers within the Wye catchment are set out in these documents:

- Compliance Assessment of Welsh River SACs against Phosphorus Targets - NRW (2021) - https://bit.ly/3jRFv8X
- River Wye SAC nutrient management plan (for England) – Environment Agency & Natural England - https://bit.ly/3FnpK2q

Phosphorus targets, or standards, for non-SAC rivers fall under the EU Water Framework Directive. The method for calculating WFD standards for phosphate in UK rivers is including in this document:

- Updated recommendations of phosphorus standards for rivers - WFD TAG (2013) - https://bit.ly/38R9iYY

Specific laboratory methods are used to analyse phosphate for the purpose of setting and assessing targets. Those assessments are annual or growing season means, taken over several years, by standard monitoring programmes. **Therefore, citizen science data (gathered using test kits such as those outlined in table 2) cannot be directly used to assess whether a river is meeting, or directly compared with, the standards set.**

## What can citizen science data tell us about phosphorus in rivers?

Lots of gaps currently exist in professional standard water quality data sets as resources are limited for monitoring. Citizen science data provides an opportunity to better understand nutrient fluxes at places or times where monitoring is not possible by regulatory bodies.

Despite it not being possible to use citizen science data to assess whether or not a river is meeting the regulatory standards set, citizen science data can help identify key problem areas and emerging issues relating to phosphorus contamination, especially when gathered alongside other water quality parameters (such as electrical conductivity, pH, nitrate and turbidity).

**The more data citizen scientists can gather using standardised procedures and equipment, the clearer the picture will be.**

## Other useful sources of information for learning

EPA information page on phosphorus: https://bit.ly/3zWCfie

Hach information sheet on phosphorus tests: https://bit.ly/3jSgPNu

| | Reactive Phosphorus | Total Phosphorus |
|---|---|---|
| **Natural Resources Wales (NRW)** | **Laboratory equipment:** Discrete analyser (Gallery Plus)<br><br>**Method:** Orthophosphate reacts with ammonium molybdate and antimony potassium tartrate under acidic conditions to form a complex which, when reduced with ascorbic acid produces an intense blue colour, the absorbance of which is measured at 880nm.<br><br>**Range:** 0-2.5 mg/l (low range)<br><br>**Reported as:** $PO_4$-P ("orthophosphate as phosphorus")<br><br>**Accreditation:** UKAS | **Laboratory equipment:** Discrete analyser (AQ2)<br><br>**Method:** Condensed and organic phosphates are digested via oxidation when the sample is mixed with by persulphate under alkaline conditions and heated to 121 +/- 2°C. Orthophosphate ions are then analysed using the same approach detailed on the left.<br><br>**Range**: 0-0.5 mg/l<br><br>**Reported as:** $PO_4$-P ("orthophosphate as phosphorus")<br><br>**Accreditation:** Not currently accredited |
| **Environment Agency (EA)** | **Laboratory equipment:** Discrete analyser<br><br>**Method:** As above<br><br>**Range:** 0-2 mg/l (normal range)<br><br>**Reported as:** $PO_4$-P ("orthophosphate as phosphorus") | *Details of specific method of analysis not available at time of writing* |
| **Dŵr Cymru Welsh Water (DCWW)** | **Laboratory equipment:** Discrete analyzer (Aquakem 600)<br><br>**Method:** LPM036 Determination of Nutrients<br><br>**Range:** 0.03 –1.2 mg/L<br><br>**Reported as:** $PO_4$-P ("orthophosphate as phosphorus")<br><br>**Accreditation:** UKAS/ DWTS | **Laboratory equipment:** ICPMS<br><br>**Method:** LPM038 Determination of Metals Total<br><br>**Range:** 0.055-2.0 mg/L<br><br>**Reported as:** $PO_4$-P ("orthophosphate as phosphorus")<br><br>**Accreditation:** UKAS/ DWTS |
| **Wye and Usk Foundation (WUF)** | **Field equipment (online analysers):**<br><br>- Hach Phosphax Sigma, Solitax probe and Sigma tax<br>- Envitech Bluemon Ortho & Total Phosphate Analyser | |

**Table 3.** Methods used to measure reactive and total phosphorus across by different organisations across the Wye catchment