

## Analysis of Phosphate

---

### OBJECTIVE

Phosphate ( $\text{PO}_4^{3-}$ ) is a natural and anthropogenic component of surface waters. It is a plant nutrient, and its presence can affect vegetation growth and thus affect aquatic habitats.

### EQUIPMENT

Hach DR/2010 spectrophotometer  
25 ml mixing graduated cylinder with stopper  
two 25 ml measuring cells for Hach 2010  
phosphate reagent set (Molybdate Reagent and Amino Acid Reagent)  
50.0 ppm  $\text{PO}_4$  standard (Hach)  
glass volumetric flask (100 or 250)  
distilled water  
water waste container  
phosphate reagent waste container  
distilled water squirt bottle  
safety equipment: Nitrile gloves, lab coat, lab glasses, fume hood

### STANDARD AND SAMPLE PREPARATION

- Remove the appropriate standard from the refrigerator several hours before measuring phosphate.
- Aliquot at least 60 ml of sample into plastic beakers and allow to reach room temperature.
- Mix phosphate standard immediately before test by adding 50 ppm standard to appropriate volumetric flask and topping with distilled water (20 ml in 100 ml or 50 ml in 250 ml).

### TEST PREPARATION

- Turn on spectrophotometer (lid closed), and allow it to self-test.
- Enter test number (type "485", then ENTER).
- Dial in wavelength if instructed by the machine to do so.

### REAGENT BLANK PREPARATION

When using a new set of reagents for the first time, run a reagent blank by using deionized water from the kit as the sample (see 2.5). Record the measured value and report it to the Lab Supervisor, who will correct all measurements made with that reagent set.

### TEST INITIATION

- Pour 25 ml sample into measuring cylinder.
- Measure 1 ml (using dropper) of Molybdate Reagent into cylinder.

## Analysis of Phosphate

---

- Measure 1 ml (using dropper) of Amino Acid Reagent into cylinder.
- Stopper cylinder and invert several times to mix.
- Start timer on spectrophotometer (SHIFT-TIMER).
- Pour 25 ml of sample into 25 ml cell (this is the sample blank).

### PHOSPHATE MEASUREMENT

- When timer sounds (after 10 minutes), wipe sides of sample blank cell and place in the machine. Blank the machine (ZERO). Always orient sample cells with writing facing to your left.
- Pour 25 ml of sample plus reagent from cylinder into second sample cell and measure (toggle with arrow key to  $\text{PO}_4$ ).
- Record concentration on appropriate data sheet.
- If sample is out of range, machine will display UR (under-range) or OR (over-range). If UR, record UR. If OR, dilute sample and start over at 2.5.a. When reading value, record value and dilution factor (e.g., if diluting in half, write "VALUE x2").
- Repeat 2.5 and 2.6 for each sample and standard. Standards should be measured at the beginning and end of the test, and after each 10 samples in between.
- You can perform simultaneous sample preparations by maintaining constant time between each sample preparation and sample reading. Rinse sample cells between samples, and shake out excess water.

### QUALITY CONTROL

Standard should read 10.0 mg/L  $\text{PO}_4$ , or within 10% of this value, after correction with reagent blank. If standard measurements deviate from this value, notify Lab Supervisor.

### DISPOSAL

After sample is measured, empty contents of blank cell into waste water container. Empty contents of cylinder and sample plus reagent cell slowly into phosphate reagent waste container, and rinse once into same waste container with distilled water. Rinse all containers again into waste water container with distilled water. Dispose of waste water container contents into toxic sink.

### PRECAUTIONS

Reagents contain sulfuric acid and other toxic components. Use the fume hood when performing these measurements.