

Development of a Paper-Based Fluidic Device for Phosphorus Detection

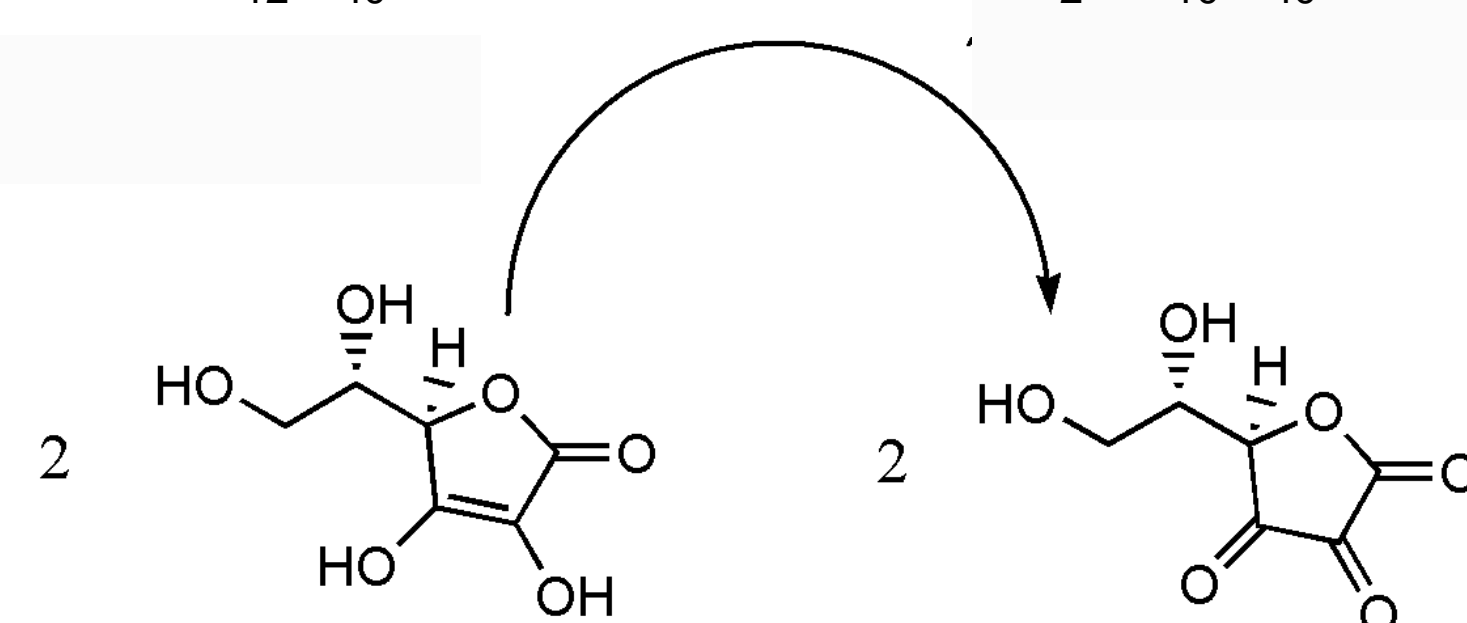
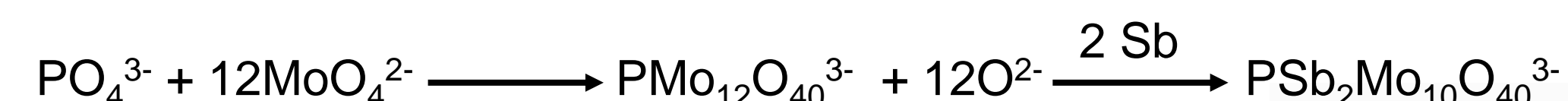
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Introduction

Phosphorus is an essential nutrient for maintaining a healthy aquatic ecosystem; however, heightened levels of phosphorus can have negative effects owed to increased plant growth. Upon decomposition of plants such as algae, oxygen levels can be depleted, resulting in anoxic zones. The ability to detect phosphorus, specifically in bodies of water, is thus necessary to evaluate environmental quality. The most common method for phosphorus detection is standard method 4500-PE in which phosphorus is detected colorimetrically through the reduction of an antimony-phosphomolybdate complex. This method, however, requires sample preservation, utilizes large volumes of reagents and requires the use of an absorbance spectrometer for quantitation. It would be advantageous to explore methods that are rapid, portable, and quantitative. Here we have developed a phosphorus detection method utilizing paper-based fluidic devices (PFDs). Phosphorus is detected as phosphate employing a modification of standard method 4500-PE which uses microliter reagent volumes. These PFDs are smaller than a business card and are simple to fabricate. Channels for reagent mixing were fabricated on cellulose filter paper using Microsoft Power Point and a Xerox ColorQube wax printer. A blue color develops upon mixing of a phosphate-containing sample and a combined reagent which consists of sulfuric acid, ammonium molybdate, potassium antimonyl tartrate, and ascorbic acid. The intensity of the blue color directly corresponds to the concentration of phosphorus which is measured using ImageJ. Detection of a concentration as little as 300 ppb phosphorus has been found. The selectivity of this phosphorus PFD is also affirmed in the presence of common ions. This method can be applied in the field and has the potential to identify areas of abnormal nutrient loading into a body of water.

Phosphorus Sensor Chemistry



Experimental

- Two- and three-channel devices (2.5 mm channel width) were fabricated using Microsoft Power Point and printed on Whatman grade 2 cellulose paper using a wax printer
- Standard method 4500-PE was modified for the paper-based fluidic device, optimizing the $\text{H}^+:\text{MoO}_4^{2-}$ ratio in the combined reagent
- An external calibration was performed using 0.050, 0.075, 0.15, 0.30, 0.50, 0.75, and 1 mg/L PO_4^{3-} -P, made from a concentrated stock solution of $\text{KH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$
- Two- and three-channel devices were loaded with 5 μL of a combined reagent and 5 μL phosphate-containing sample in the left and right channels, respectively
- Data for the external calibration curve was generated through quantitation of color density using ImageJ
- A selectivity study was performed against 1 ppm concentrations of Cl^- , F^- , NO_3^- , NO_2^- , and NH_4^+

Combined Reagent Recipes Tested

Reagent	CR - I	CR - II	CR - III	CR - IV	CR - V	CR - VI
126 mM AM	40 μL	40 μL	40 μL	40 μL	40 μL	40 μL
11.4 mM PAT	60 μL	80 μL	100 μL	100 μL	100 μL	80 μL
5 N H_2SO_4	300 μL	300 μL	300 μL	300 μL	300 μL	300 μL
0.5 M AA	100 μL	100 μL	100 μL	160 μL	260 μL	260 μL

The above combined reagents were tested in comparison to Combined Reagent 3* below. All further analyses reported employed Combined Reagent 3* due to optimal color formation.

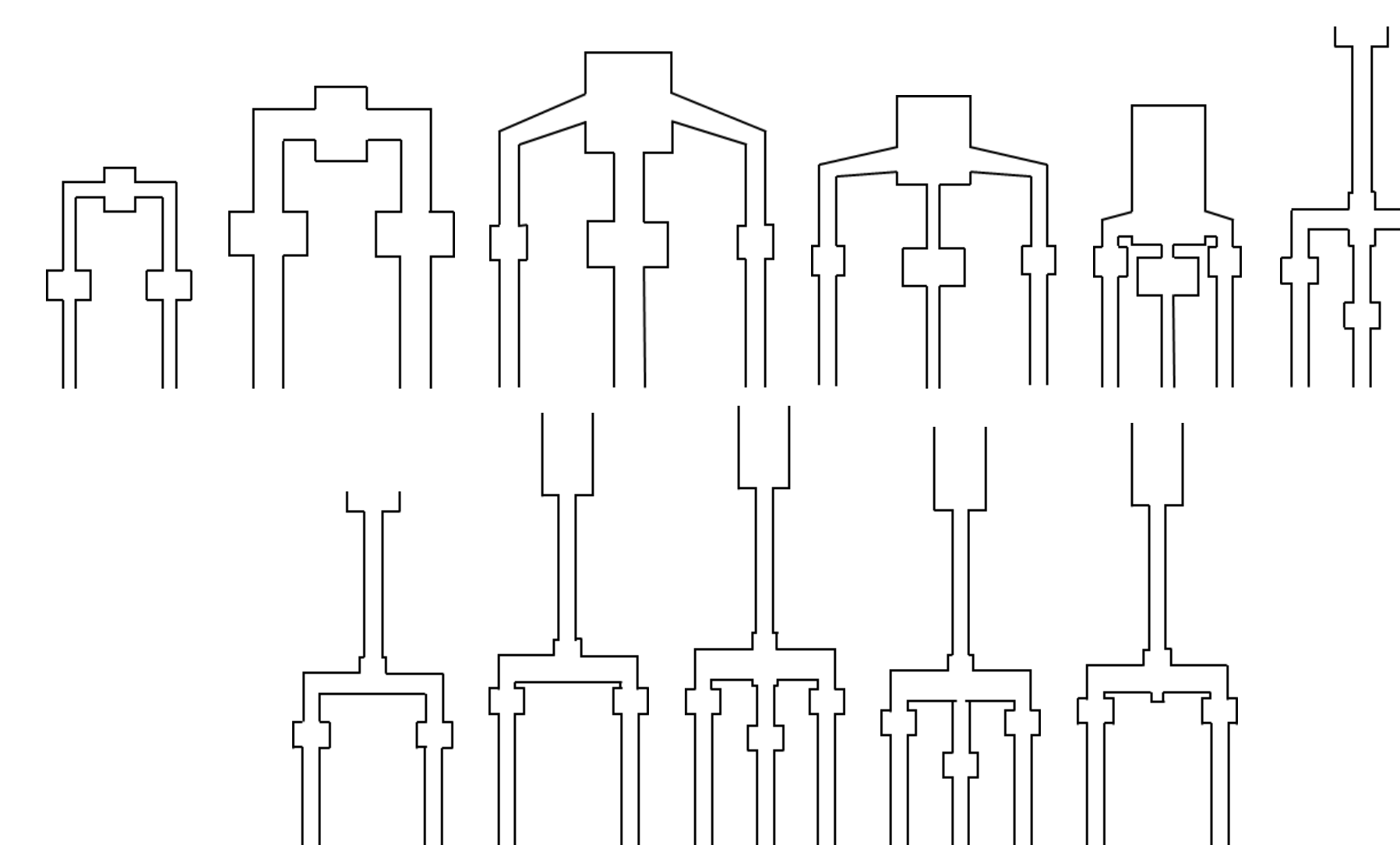
Reagent	CR 1	CR 2	CR 3*	CR 4	CR 5	CR 6	CR 3b*	CR 3c	CR 3d
126 mM AM	15 μL	15 μL	40 μL	40 μL	40 μL	40 μL	40 μL	80 μL	60 μL
11.4 mM PAT	2.5 μL	0 μL	50 μL	0 μL	70 μL	30 μL	50 μL	50 μL	50 μL
5 N H_2SO_4	52.5 μL	52.5 μL	100 μL	100 μL	80 μL	120 μL	120 μL	120 μL	120 μL
0.5 M AA	30 μL	30 μL	80 μL	80 μL	80 μL	80 μL	60 μL	80 μL	80 μL
H_2O	0 μL	2.5 μL	0 μL	50 μL	0 μL	0 μL	0 μL	0 μL	0 μL

*Combined Reagents 3 and 3b resulted in the most intense blue color in the presence of phosphate, while minimizing non-specific color development

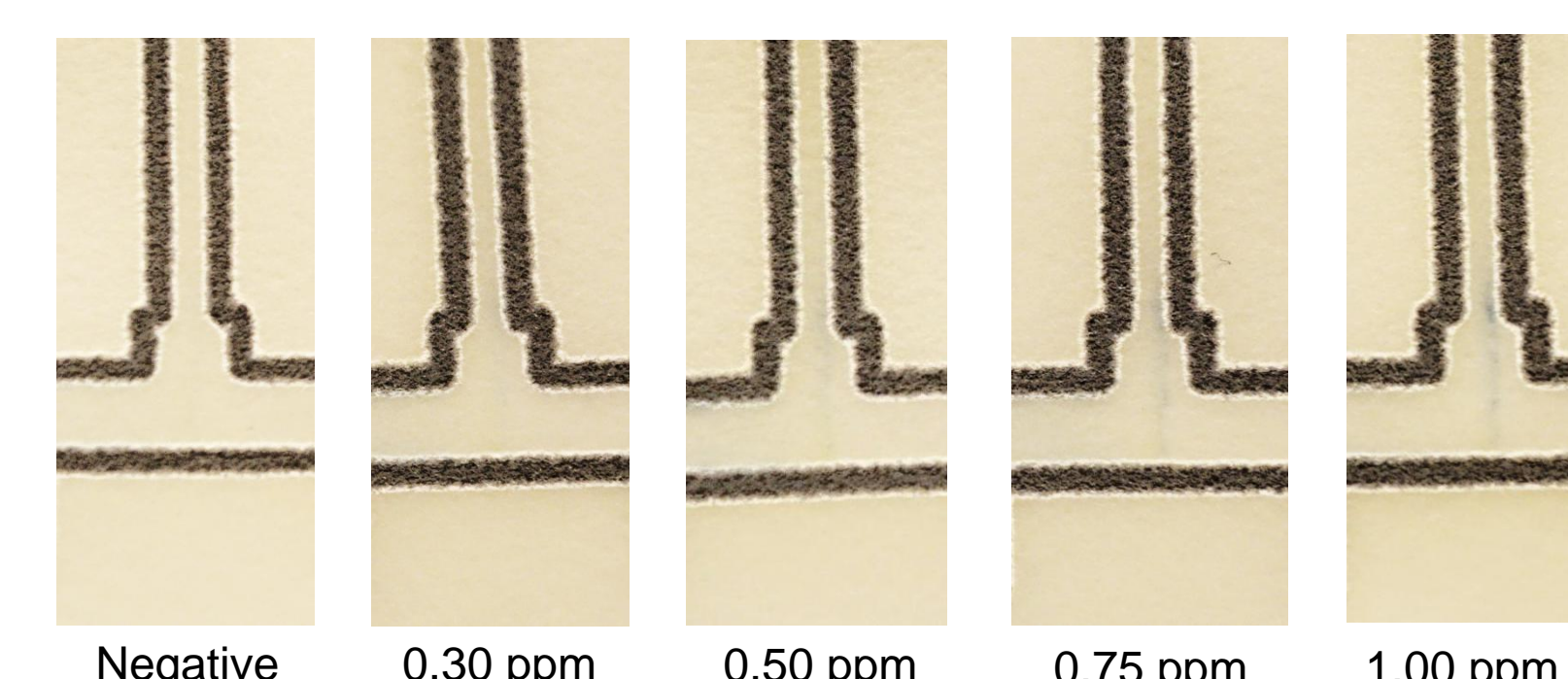
CR = Combined reagent AM = Ammonium molybdate
PAT = Potassium antimonyl tartrate AA = Ascorbic acid

Results

PFD Designs Tested

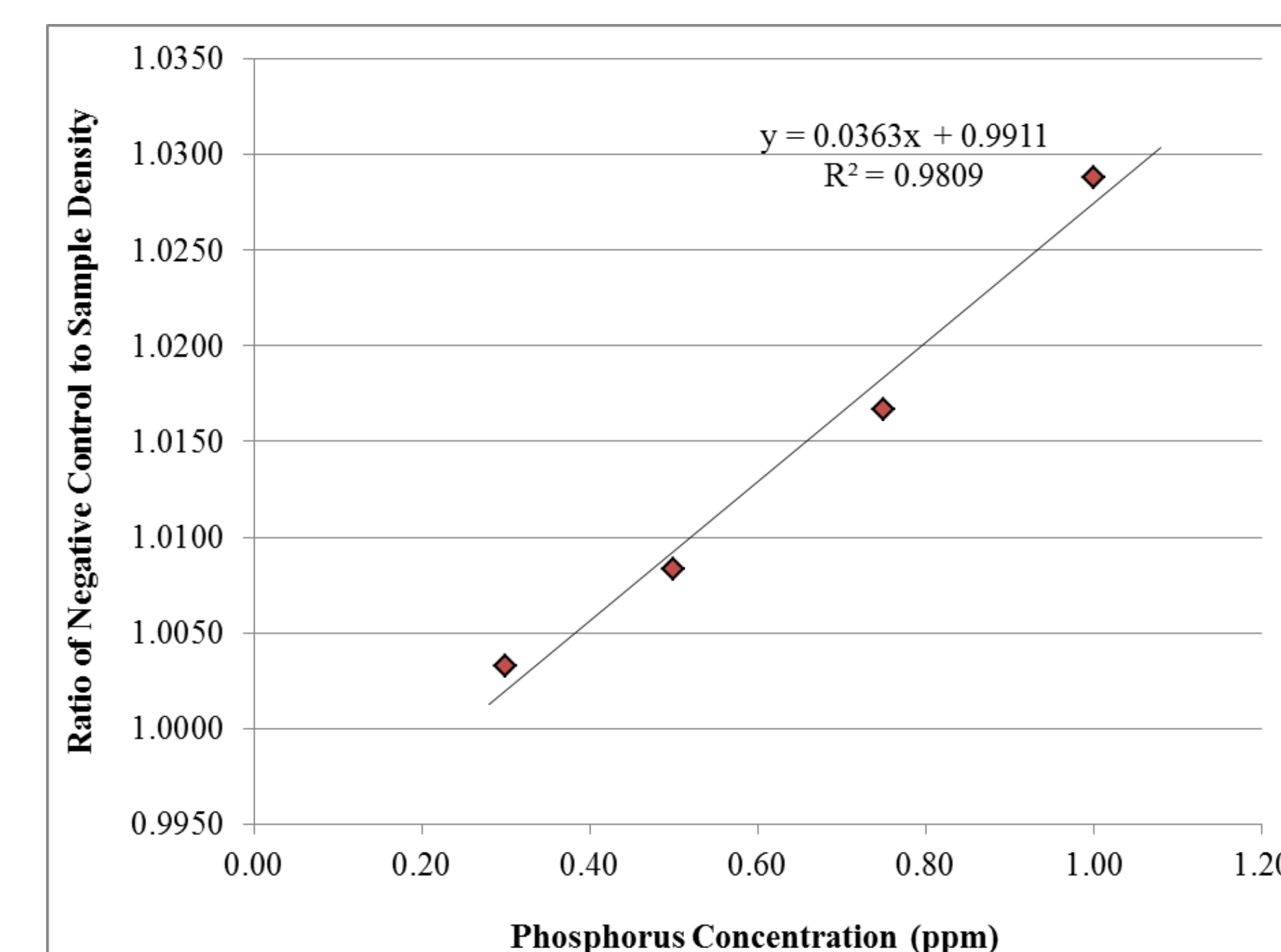


External Calibration Curve PFDs

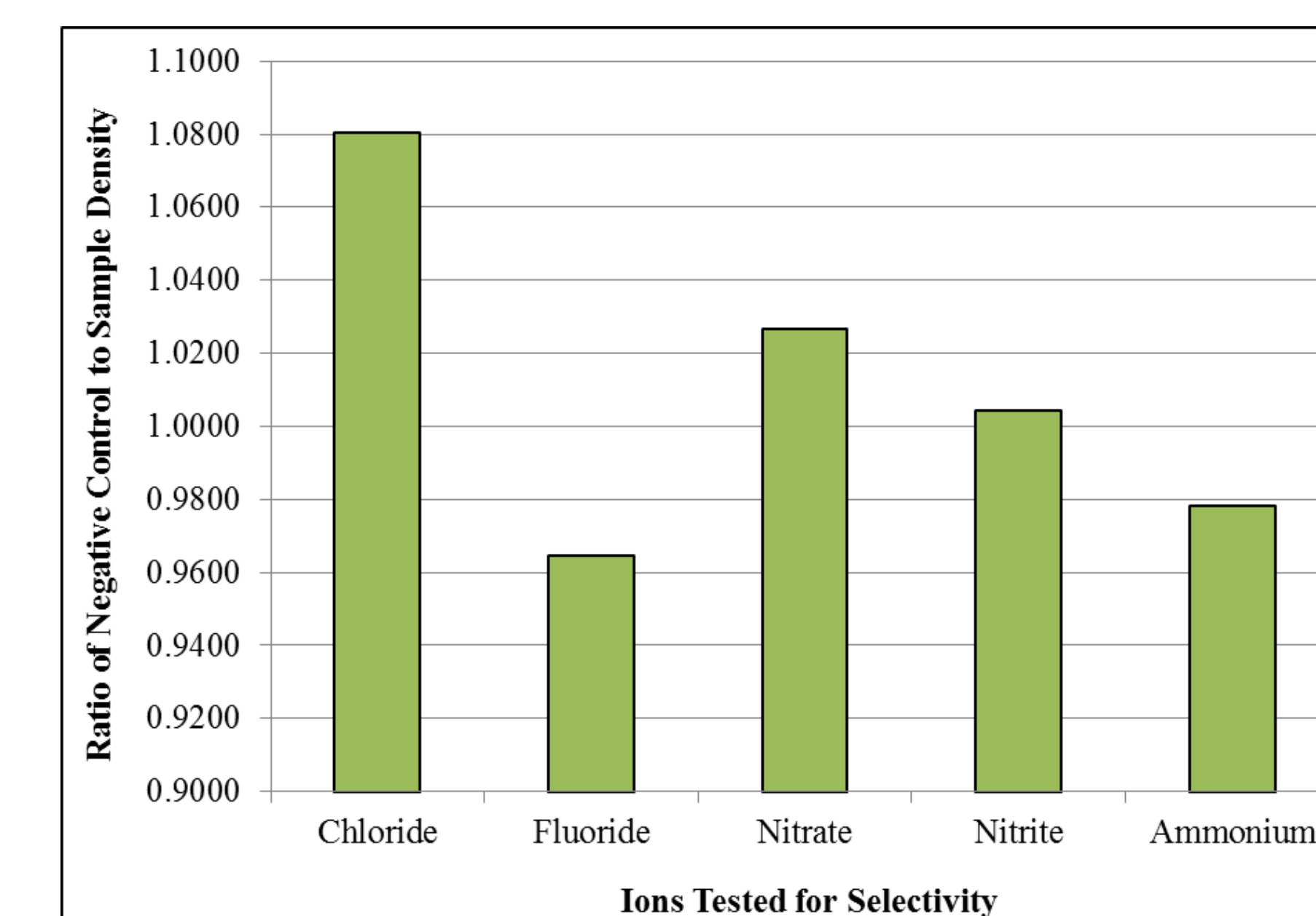


Water drives the phosphate-containing sample and the combined reagent vertically through the channels until mixing is completed producing a blue color (Antimony-Phosphomolybdenum Blue) in the presence of phosphate

External Standard Calibration Curve



Selectivity Study



Conclusions

- The phosphorus PFD provides for a portable detection device that can be employed on-site
- Phosphorus can be detected as low as 300 ppb on PFDs using microliter reagent volumes
- ImageJ analysis reveals that the chloride, nitrate, and nitrite are potentially interfering ions, although visual inspection of the devices did not reveal blue color formation
- Different devices have different hues upon wetting, possibly interfering with density analysis. Future devices will incorporate an internal standard to correct for these color differences
- In the future, a cellular phone application will be developed to allow for on-site quantitation

References

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- Rice, Eugene W., Roger B. Baird, Andrew D. Eaton, Lenore S. Clesceri, eds. "Standard Methods for the Examination of Water and Wastewater", 22nd Edition (2012) American Public Health Association
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