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Application Note 39420813

Automated In-line Digestion and Analysis of Total Phosphorous (TP) and Total Nitrogen (TN) in Environmental Water Samples

Keywords
Alkaline Persulfate Digestion
TKN – Total Kjeldahl Nitrogen
TN – Total Nitrogen
TP – Total Phosphorous

Introduction

A high proportion of the workload in environmental laboratories involves analysis of total phosphorous (TP) and total nitrogen (TN) in water samples for Clean Water Act compliance reporting. At the present time, these analyses must be performed using manual or semi-automated methods as there are no U.S. EPA-approved automated methods.

The U.S. EPA parameter for total nitrogen is the sum of Total Kjeldahl Nitrogen (TKN) and Nitrate plus Nitrite Nitrogen. Water samples that have undergone Kjeldahl digestion can be analyzed for total phosphorous as well as TKN which only partially mitigates sample handling and workload issues. Fully automated simultaneous analysis of TP and TN cannot be achieved because an approved method for determining TN as a single analyte is not available.

This study examined the use of an alkaline persulfate digestion as an alternative to Kjeldahl digestion to support analysis of both total nitrogen and total phosphorous. Analytical results obtained from automated in-line digestion and simultaneous analysis of TP and TN are presented and contrasted with results obtained by currently approved manual and semi-automated methods.

Alkaline Persulfate Digestion Chemistry

Total Nitrogen

Alkaline persulfate oxidation of TN is a common method in oceanographic chemistry. Samples are digested using persulfate at a high pH to convert all nitrogen to nitrate. Any technique capable of determining nitrate can be used for final measurement.

$$(NH_2)_2CO + 8\ S_2O_6^{-2} + 18\ OH^- \longrightarrow 2\ NO_3^- + CO_2 + 16\ SO_4^{-2} + 11\ H_2O$$

There are no significant interferences with the method if the peroxydisulfate concentration is high enough. Because the digestion is under alkaline conditions, chloride does not interfere. Some organic nitrogen compounds may not be completely recovered. Also, samples with high organic loads, or high TSS loads may consume persulfate.

Total Phosphorous

Alkaline persulfate oxidation of TP using the same digestion described above for TN enables the analysis of low concentrations of TN and TP from a single digestion. The method is not U.S. EPA approved for either TN or TP at this time. The sample is digested in a tube, on a block or in an autoclave in the presence of persulfate at a high pH converting all nitrogen to nitrate, and all phosphorous compounds to orthophosphate.

Presented at the 2013 Pittsburgh Conference on Analytical Chemistry and Applied Spectroscopy, Philadelphia, Pennsylvania, March 17-21, 2013

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Experimental

Manual Digestion

The total phosphorus concentration was determined by an OI Analytical method (PN 327559) Low Level Phosphorus - All Forms, USEPA by Flow Injection Analysis (FIA)[1], a modification to U.S. EPA method 365.1[2] that maintains the acidity and acid to molybdate ratio described by Murphy and Riley.[3] The manual acid digestion described in the EPA method was scaled down proportionally to 5 milliliter sample volumes and digested using 16 x 100 mm Teflon® lined screw cap vials. Non-acidified standards and samples were digested at 121 °C for 40 minutes using a bench top thermo reactor similar to the one pictured in Figure 1.

Figure 1. Thermo Reactor for Manual Digestion of Total Nitrogen and Phosphorus

Total phosphorus and total nitrogen were manually digested using the alkaline persulfate reagents specified in U.S. Geological Survey Water – Resource Investigative Report 03-4174[4] and OI method (PN 327569) Total Nitrogen by Manual Alkaline Persulfate Digestion and Segmented Flow Analysis.[5] The digestion was modified by scaling down sample volumes and digesting in 16 x 100 mm Teflon ® lined screw cap vials on the thermo reactor. Recovery of nitrogen and phosphorus by alkaline persulfate digestion depends on the progressive decrease in pH from about 12 to about 2. Under alkaline conditions the persulfate oxidizes reduced nitrogen compounds to nitrate. As the persulfate decomposes it becomes bisulfate ion. Once all of the persulfate is converted to bisulfate the solution becomes acidic. The low pH converts phosphorus compounds to orthophosphate.

Total phosphorus in the alkaline digestates was determined using the modified version of EPA 365.1 in OI Analytical method (PN 327559)[1] that was used to determine total phosphorus in acid digestates. Total Nitrogen was determined as nitrate according to the OI Analytical method (PN 327569) Total Nitrogen by Manual Alkaline Persulfate Digestion and Segmented Flow Analysis. The nitrate determinative step can be considered equivalent to EPA 353.2.[6]

All total phosphorus and total nitrogen analyses of acid and alkaline digests were performed on an OI Analytical FS 3100 Continuous Flow Analyzer.

Automated Digestion

An OI Analytical FS 3100 Continuous Flow Analyzer was configured for in-line digestion of total nitrogen and total phosphorus by adding a heating coil and UV irradiation module. The sample is mixed with sodium persulfate

reagent, either acidic or alkaline and passed through the heater followed by UV radiation at 254 nm. The persulfate is catalyzed creating a strongly oxidizing solution that converts phosphorus compounds to phosphate and, under alkaline conditions, converts nitrogen compounds to nitrate. The phosphate and the nitrate reaction products were measured by EPA 365.1 and EPA 353.2 respectively.

Results and Discussion

Determination of Different Nitrogen and Phosphorus Containing Compounds

The set of environmentally important indicator compounds chosen to evaluate the effectiveness of each digestion technique are listed in Table 1.

Table 1. Indicator Compounds Used to Evaluate Digestion Efficiency of TN and TP Methods

Compound	Target Analyte	Organic or Inorganic
Nitrate Nitrogen*	NO_3^-	Inorganic N
EDTA	R-N-R	Organic N
Urea	$R\text{-}NH_2$	Organic N
Glycine	$R\text{-}NH_2$	Organic N
Niacin	R-N-R (ring)	Organic N
Ammonium Nitrogen	NH_4^+	Inorganic N
Nitrite Nitrogen	NO_2^-	Inorganic N
Phosphate*	PO_4^{-3}	Inorganic P
Tripolyphosphate	P-O-P	Inorganic P
Hexametaphosphate	P-O-P	Inorganic P
Pyrophosphate	P-O-P	Inorganic P
Trimethylphosphate	$(RO)_3OP$	Organic P

*chemical form determined by the method

Each compound was prepared as an individual standard at 1 mg/L in reagent water and then digested. Results were compared to calibration curves prepared from a series of calibration standards carried through the same digestion procedure.

Total Phosphorus

The digestion efficiency of the alkaline digestion for total phosphorus was compared with the efficiency of the approved acid digestion method describe in EPA 365.1, (results shown in Figure 2). Trimethylphosphate (organic phosphorus) and phosphate were completely recovered with an alkaline digestion time of 1 hour. Hydrolysable phosphorus compounds, (P-O-P bonds), were not quantitatively recovered until the digestion time was extended to 3 hours.

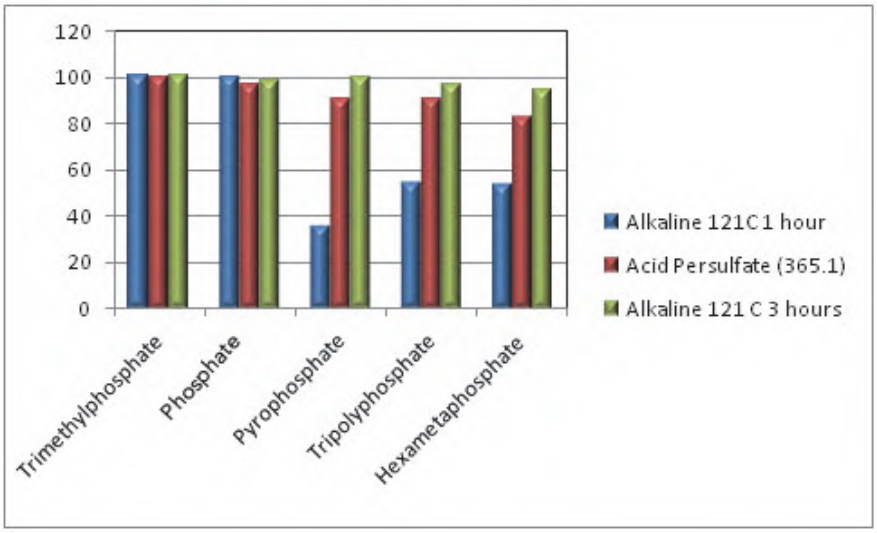

Figure 2. Recovery of Phosphorus Compounds Obtained by Manual Acid and Alkaline Persulfate Digestion

Seven wastewater matrices were analyzed for total phosphorus by the 3-hour alkaline persulfate digestion and compared to the U.S. EPA approved method. Results presented in Figure 3 indicate that the two digestion methods are equivalent. Because 40 CFR part 136.6 allows the interchange of oxidant, the alkaline digestion should be considered a U.S. EPA approved alternative digestion for the determination of total phosphorus by EPA 365.1.

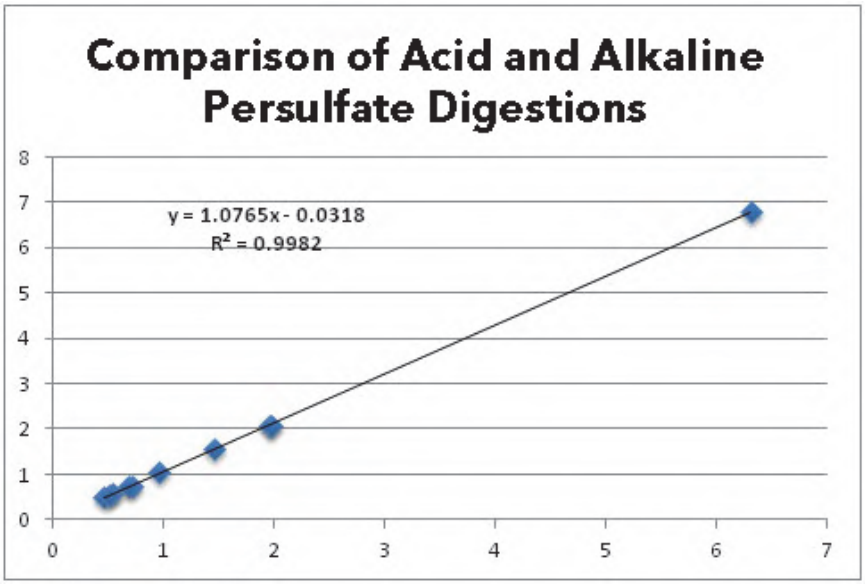

Figure 3. Scatter Plot Comparison of Total Phosphorus Results Obtained by Alkaline and Acid Persulfate Digestion

Total phosphorus analysis was attempted by in-line, UV-heated acid persulfate digestion (results are shown in Figure 4). The acceptable recovery of organic phosphorus and orthophosphate along with very low recovery of hydrolysable phosphorus are consistent with the long digestion time required for quantitative recovery using manual methods.

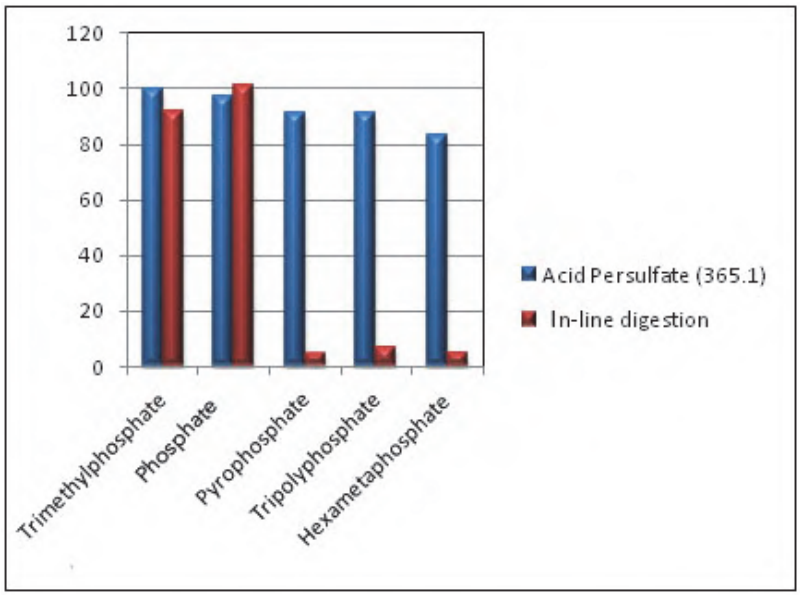

Figure 4. Automated Digestion Recovery of Total Phosphorus Compared to 365.1

Total Nitrogen

While we expected that nitrogen would not require a three hour digestion, our intent was to measure total nitrogen and total phosphorus from a single digestion. Therefore, a three hour alkaline persulfate digestion was used to manually digest total nitrogen. Recoveries for the manual alkaline digestion of total nitrogen are presented in Figure 5.

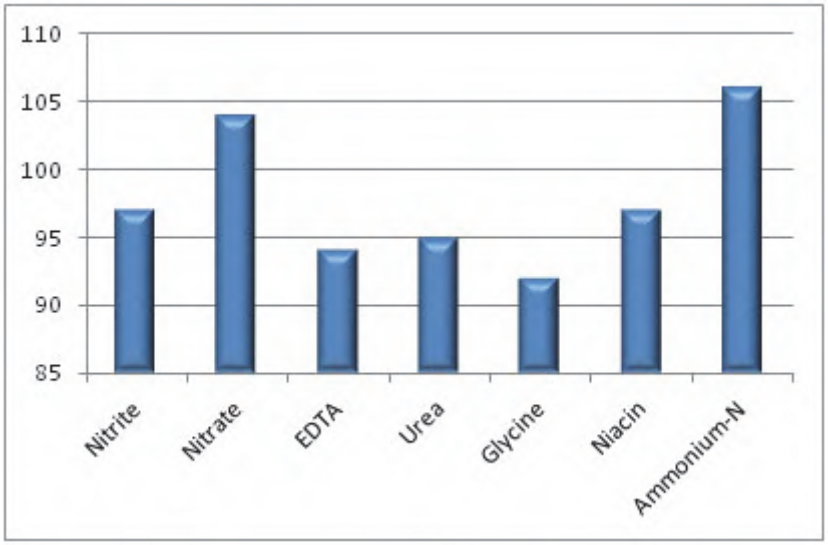

Figure 5. Recovery of Nitrogen Compounds Obtained by Manual Alkaline Digestion

Once recoveries were established for the manual alkaline digestion of total nitrogen, total nitrogen was then analyzed by the in-line, UV-heated persulfate digestion method. Figure 6 is a summary of the results obtained.

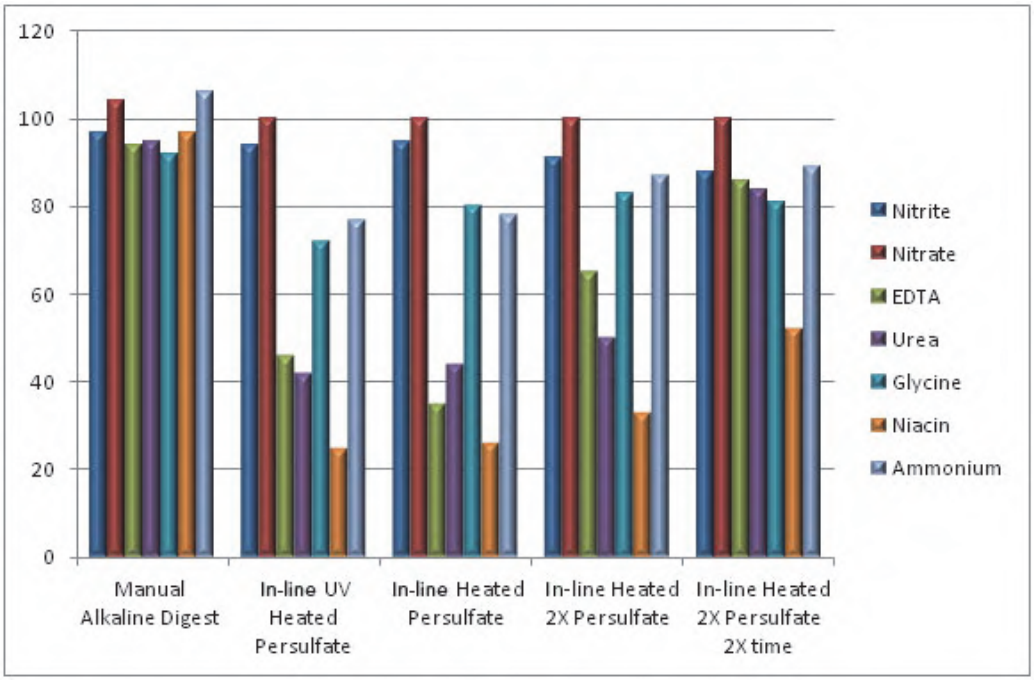

Figure 6. On-line Digestion of Total Nitrogen

As shown in Figure 6, UV irradiation has no affect on the recovery of total nitrogen. UV irradiation was not used in the subsequent experiments. Extra persulfate was added with limited success. As with total phosphorus, digestion time is the most important variable in the quantitative recovery of total nitrogen.

Summary and Conclusions

A manual alkaline persulfate digestion carried out for 3 hours at 121 °C in 16 x 100 mm vials using a bench top thermo reactor followed by analysis by EPA-approved methods 352.2 and 365.1 can be used to quantitatively recover total nitrogen and total phosphorus respectively from a single digestion. Alkaline digestion results for total phosphorus can be considered equivalent of EPA approved Method 365.1.

In-line digestion methods are time dependent and require complete validation of the digestion conditions, specifically digestion time. Comparison of recoveries between in-line methods and a manual method should be made prior to reporting results. In-line digestion of hydrolysable phosphorus was incomplete.

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6. EPA Method 353.2 Ver. 2.0, Determination of Nitrate-Nitrite Nitrogen by Automated Colorimetry, August 1993.

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Publication 39420813